

**ELF Communications System
Ecological Monitoring Program:
Soil Amoeba – Final Report**

R. Neal Band

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13. ABSTRACT (Maximum 200 words) The U.S. Navy has completed a program that monitored biota and ecological relationships for possible effects from electromagnetic (EM) fields produced by its Extremely Low Frequency (ELF) Communications System. This report documents the results and conclusions of soil amoeba studies conducted near the Navy's transmitting antenna in Michigan. From 1982 through 1994 researchers from the Michigan State University (MSU) monitored species and populations of soil amoebae found in ELF system area soils. Six variables characterizing population size, activity, and diversity, as well as genetic heterogeneity and growth, were examined in areas near (treatments) and far (control) from the antenna. The research team also measured ambient soil factors such as temperature, moisture, and nutrient chemistry. Data were analyzed using analysis of variance and BACI techniques. Statistical analyses of diversity, genetic heterogeneity, and growth indicated no EM effects on these variables. Results on seasonal averages, peak numbers, and encystation activity were mixed; however, the number of significant differences was small and the overall pattern was not related to EM exposure. The principal investigator concluded no effects to amoebae from exposure to EM fields produced by the ELF system.							
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FOREWORD

This report by researchers from Michigan State University (MSU) summarizes the results and conclusions of their study of soil amoebae. In this effort, MSU monitored species of soil amoebae exposed to electromagnetic fields produced by the U.S. Navy's ELF Communications System in Michigan. The Space and Naval Warfare Systems Command (SPAWAR) funded this MSU study through contracts N00039-81-C-0357, N00039-84-C-0070, N00039-88-C-0065, and N00039-93-C-0001 to IIT Research Institute (IITRI). IITRI, a not-for-profit organization, provided engineering support to MSU and managed their study through subcontract agreements.

MSU initiated their studies in late 1982. Their early efforts focused on selecting study sites, validating assumptions made in proposals, and characterizing critical study aspects. As these tasks were accomplished in 1983 and 1984, MSU then emphasized accumulating a data base through 1993. The MSU research team and IITRI evaluated each study variable for continued funding before contract renewals in 1984, 1988, and 1993. As a result, several originally proposed study elements were either expanded or discontinued in subsequent periods of performance.

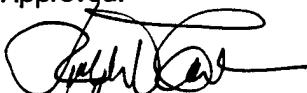
Since its inception, scientific peers have reviewed the technical quality of this study on an annual basis. In similar fashion, a draft of this report has been reviewed by peers with experience in soil microbiology, cell biology, statistics, and electromagnetics. MSU authors have considered, and addressed, peer critiques before submitting their revised manuscript to IITRI. Except for added prefatory and title pages, MSU's manuscript is here issued by IITRI on behalf of SPAWAR without further changes or editing by IITRI or SPAWAR.

Respectfully submitted,
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GLOSSARY

allele: one of several forms (isoforms) of a gene at a given point (locus) on a chromosome. In diploid organisms chromosomes are found in pairs so that two alleles are found at each locus on the chromosome pair and can be separated by migration in an electric field (electrophoresis) and visualized by staining techniques.

allozyme: isoforms of an enzyme produced by different alleles that use the same substrate but have different charges and sizes. Different alleles at a locus produce closely related enzymes while isoforms produced by different gene clusters (i.e. loci) are sufficiently different to be detected and separated by electrophoresis.

clone: population of cells identical with a single ancestral cell.

cyst: dormant amoeba resistant to starvation.

exponential growth: growth of cells with a constant doubling time.

LSS: low salt saline.

study sites:

CON, control

ANT, antenna

GND, ground wire

ORG, upper organic soil horizon

MIN, lower mineral soil horizon

soil enrichment: culture method to selectively support considerable growth of a group of microorganisms to

facilitate identification and isolation. For protozoa feeding on bacteria, enrichment consists of adding an excess of bacterial food to support a bloom of organisms.

vegetative amoeba: actively growing organisms as opposed to dormant cysts.

ABSTRACT

The present study was designed to detect possible effects on populations of soil amoebae by the extremely low frequency electromagnetic emissions (ELF-EM) from the Navy's ELF antenna, located in Michigan's Upper Peninsula. The studies were started before the ELF antenna was fully constructed (1983 to 1985), continuing into partial operation (1986 to 1988) and then into full operation in 1989.

Sites located adjacent to the antenna and the ground wire were used in conjunction with a control site some distance from the antenna. The sites were characterized by IITRI personnel to insure that all sites had a similar 60 Hz electromagnetic background while the control site had at least an order of magnitude lower ELF-EM intensity.

Populations of amoebae increased over the growing season, usually peaking in August/September, and then decreased in the Fall. Peak population densities appeared to be correlated with annual rainfall and soil moisture. However, no difference in population size between antenna, ground wire, and control sites was detected before or after the antenna became operational. A statistical comparison of all pre-operational versus all post-operational data for maximum population densities revealed a statistically significant, small difference between the control site and the ground site. This same method of analysis failed to reveal statistically significant differences between the control and antenna sites, and between the antenna and ground sites.

Some differences in soil chemistry were noted between sites and years, but these did not relate to amoeba populations. Soil pH ranged from pH 6.0 to 7.0 at the sites and dates. Soil temperature readings fluctuated over the growing season from 10 to 20 °C.

Growth rates of Acanthamoeba polyphaga exposed to ELF-EM fields did not differ between sites. Genetic diversity studies conducted in 1986 to 1988, 1991, and 1993 failed to reveal differences between study sites.

SUMMARY

Sites: The effects of the 76 Hz extremely low frequency (ELF) electromagnetic fields generated by the U.S. Navy's ELF antenna on soil amoebae were studied from 1983 through 1993 at three sites in Dickinson County, Michigan. The three sites consisted of a control site that was 15 km from the ground site, a ground site 39 m from the overhead feed for the ground for the southern end of the north-south leg of the antenna, and an antenna site located within 30-50 m of the north-south leg of the antenna. All three sites were matched for 60 Hz electric field and magnetic flux density exposure prior to operation of the antenna. Exposure of soil amoebae to 76 Hz generated electromagnetic fields started in 1986 and continued after the Michigan transmitter went to full power in 1989. The amoebae at the antenna site were exposed to the greatest electromagnetic field; those at the ground were exposed to intermediate levels; and those at the control site were exposed to very low levels.

Soil amoeba studies included three primary types of data collection: (1) total soil amoeba counts and percent amoebae that were encysted; (2) growth of *Acanthamoeba polyphaga* clones in the field with special culture vessels designed to expose amoebae to ELF-EM fields; and (3) analysis of isoenzyme patterns for clones of *A. polyphaga* as a means of determining possible effects of the ELF antenna on genetic heterogeneity.

Amoeba population density: Soil amoeba population densities varied from 1000 to 2.3 million per gram of soil. No consistent, significant site differences in total population size were

detected using an analysis of variance ($p < 0.05$) for pre-operational years (1984, 1985), testing (at less than full power in 1986, 1987 and 1988), or operational years (1989 to date). A statistical comparison of all maximum population densities before and after the antenna became operation did reveal a small difference between the control site and the ground site. Using this same analysis, no differences were detected between the control and antenna sites or between the antenna and the ground sites. These results indicated that ELF electromagnetic fields had no detectable effect on population densities of soil amoebae. Percentage of soil amoebae encysted, perhaps a measure of population stress, did not correlate well with high or low counts at the sites. Total soil amoeba counts at all sites were more variable from sampling period to sampling period than between sites. The largest soil amoeba populations occurred in either July or August in most years with maximum numbers varying from 2000 to 4000 amoebae per gram of mineral soil in dry years such as in 1986 to greater than two million amoebae per gram of organic soil at some sites in wet years. Organic horizons consistently supported larger populations than did mineral horizons at all study sites. Periods with high or low counts in the organic horizons corresponded to periods of high or low counts in the mineral horizons.

For each site, amoeba numbers were related to soil moisture in the organic horizon but a similar correlation could not be detected in the lower, mineral horizon. Soil amoebae achieved greater population densities in years with more than average

rainfall and lowest densities in years with less than average rainfall, again indicating the importance of soil moisture. Soil temperature readings were similar at all sites and varied over a fairly narrow range of 6 to 18 °C over the sampling period from June to October and readings were generally between 12 and 16 °C in the period from July to September when maximum population numbers were reached.

Chemical analyses showed that the site soils were fairly well matched. Differences between sites and years did occur particularly for soil PO₄ and organic nitrogen; however, these differences were not consistent from year to year. With rare exception, soil amoeba counts did not differ between the sites, for a given sampling date and soil horizon, with rare exception.

Soil amoebae are micropredators, and variations in total counts are thought to reflect differences in quality and quantity of food available, especially bacteria. Some bacteria are food while others are toxic. Attempts to characterize numbers of bacteria available to amoebae in the soil, using a modification of the acridine orange direct counting technique demonstrated that soil could contain as many as 10⁹ bacteria per gram of soil. However, numbers were highly variable, so attempts to use this technique routinely to explain variance in soil amoeba numbers were discontinued.

Extraction of soil DNA as an indirect estimate of bacterial biomass proved to be too variable to be of use; soil DNA could not be quantitatively separated from interfering substances in soil.

Fungi and actinomycetes were also examined. About 300 isolates were obtained from the study sites. Most were eaten or ignored by amoebae, but one of the actinomycetes (*Streptomyces* sp.) proved to be quite toxic to soil amoebae. Soil amoeba population size was related positively to their food (bacteria and fungi) and negatively to species that have developed chemical defenses against them.

Growth rate in soil cultures: *In situ* growth of *A. polyphaga* was studied using soil culture vessels designed to match ELF-EM exposures in the soil. Vessels were buried so as to be in equilibrium with the temperature of the soil. Amoebae were fed *Escherichia coli* at a concentration of 1 mg/ml, and their growth was compared across sites and years (1989, 1990 and 1991). Growth rates were determined from direct counts of organisms at the study sites. Clone isolates of *A. polyphaga* were also isolated each year. Isoenzyme analysis of the clones before and after exposure in the culture vessels was used to determine if *A. polyphaga* from the surrounding soil had contaminated the cultures. Other protozoan contaminants could be detected microscopically.

No significant difference ($p > 0.05$) in growth rates of amoebae were detected between the sites in any of the three years of the study indicating that a fully operational ELF system had no effect on growth of the amoebae.

Allozyme study: In a preliminary study of genetic heterogeneity of *A. polyphaga*, isoenzyme analyses were done with 5 clone isolates from each site in 1985, using 3 enzymes and 10

loci (Jacobson and Band, 1987). In 1986, 1987, 1988, and 1991, 10 clones from each site were used with 8 to 15 enzymes at 27 to 34 loci. In 1993, 30 clones from each site were examined with 9 enzymes at 28 loci. Calculated genetic heterogeneity was greater in 1985 (Jacobson and Band, 1987). The smaller genetic heterogeneity observed in 1986 and later years may be the result of using larger sample sizes and a greater number of genetic loci for analysis after 1985. There were no significant ($p < 0.05$) differences in genetic heterogeneity between the three sites during low-amperage (4-6 A) testing of the antenna in 1986, 15 A testing in 1987, and 75 A testing in 1988, nor during full operation in 1991 and 1993.

SUMMARY REPORT

The project objective was to examine for possible effects of ELF electromagnetic radiation from the antenna on soil amoebae. The treatment sites chosen for this study were adjacent to the Michigan ELF antenna and ground wire. A single control site was located 15 km south of the antenna.

1. STUDY SITE CHARACTERISTICS

Antenna, ground and control sites were used in this study. They were located in sugar maple (*Acer saccharum* Marshall) dominant hardwood forests, with some bass wood (*Tilia americana* L.). The soil was a sharply stratified loam, consisting of an upper organic horizon of mostly plant litter and a lower, mineral horizon of sandy loam. This section of the report describes, for the three sites, the physical and chemical characteristics that relate to growth of soil amoebae.

1.1 *Location.* The sites were selected in cooperation with IITRI personnel so that all sites had a similar 60 Hz electromagnetic (EM) background while the control site had at least an order of magnitude lower ELF (76 Hz) EM exposure than the treatment sites. The methods are given in Haradem *et al.* (1994). The data from Haradem *et al.* (1994) are summarized in Table 1, together with electric field measurements in culture vessels performed by Michigan State University (MSU) personnel (used in Section 4, *In situ*). A map of site locations is included in this report, with individual site maps (Appendix Fig. A-1).

The 20 m x 20 m sites were located as follows:

TABLE 1. 76 Hz EM average intensities at soil amoeba sites*

YEAR	Electric Field (mV/M)						Magnetic Field (mG)					
	soil			Cult			Soil			Cult		
	ANT	GND	CON	ANT	GND	CON	ANT	GND	CON	ANT	GND	CON
1985	0.00	0.00	0.00	-	-	-	0.00	0.00	0.00	-	-	-
1986	1.40 ±0.10	1.16 ±0.40	0.03	-	-	-	0.23 ±0.10	0.08 ±0.01	<.001	-	-	-
1987	5.70 ±4.00	9.90 ±5.50	0.07	-	-	-	0.84 ±3.00	0.30 ±0.05	<.001	-	-	-
1988	23.00 ±2.80	20.00 ±15.0	0.36	-	-	-	4.00 ±1.30	1.48 ±0.22	<.002	-	-	-
1989	50.50 ±4.90	35.90 ±12.0	1.37	43.66 ±10.77	23.00 ±6.67	0.99 ±0.39	8.20 ±2.70	3.37 ±0.53	.004	8.20 ±2.70	3.37 ±0.53	.004
1990	52.50 ±0.70	37.20 ±8.00	0.76	50.33 ±3.97	21.44 ±2.46	1.65 ±0.31	8.20 ±2.80	2.98 ±0.48	.004	8.20 ±2.80	2.98 ±0.48	.004
1991	52.00 ±1.40	34.00 ±8.00	0.90	50.22 ±4.38	22.66 ±1.00	1.56 ±0.70	7.15 ±2.60	2.85 ±0.40	.002	7.15 ±2.60	2.85 ±0.40	.002
1992	58.00 ±1.40	39.20 ±8.70	0.94	-	-	-	8.10 ±2.60	3.00 ±0.40	.005	-	-	-
1993	49.50 ±5.00	37.30 ±7.40	1.02	-	-	-	7.70 ±2.60	2.90 ±0.40	.005	-	-	-

*Abbreviations: Antenna (ANT); Ground (GND); Control (CON); culture vessel measurements (CULT). All reported measurements were done by IITRI except Electric Field CULT experiments which were done by MSU in 1989 to 1991 (n=9/site/yr), using electrodes spaced 1 m apart. For IITRI methods see Haradem et al., 1994; their reported measurements were: ANT site n = 2; GND site n = 6; CON site n = 1/yr.

1. Antenna (ANT) site: located within 30 to 50 m of the north/south leg of the antenna at Tier 43N, Range 20W, Sect. 23 (Appendix Fig. A-3).

2. Ground (GND) site: located 39 m from the overhead feed for the ground wire at Tier 42N, Range 29W, Sect. 11 (Appendix Fig. A-4).

3. Control (CON) site: located 15 km south of the ground site at Tier 41N, Range 29W, Sect. 21 (Appendix Fig. A-2).

1.2 Materials and Methods. Soil at the sites was a sharply stratified sandy loam. The upper "organic" (ORG) horizon consisted of decomposing plant and animal litter approximately 3 to 6 cm thick. It was sharply separated from the lower "mineral" (MIN) horizon, which was a sandy loam soil.

Soil pH, bulk density, and soil moisture content were done by the methods given in Richards (1954). Soil moisture was determined gravimetrically with soil dried to a constant weight at 105° C. In addition, soil suction (Baver et al., 1972) was determined with a pressure membrane extractor (Soilmoisture Equip. Corp., Santa Barbara, CA). Soil temperature was recorded at 4 hr intervals in the interface between soil horizons with Datapod Model DP222 data loggers (Omnidata Int., Logan, UT). Three data loggers were used at each site in the event that one or two ceased to operate. Means were used when more than one data logger functioned.

Soil chemistry, performed by MSU's Soil Testing Laboratory, was done on ORG and MIN horizons. Each sample to be tested came from 20 pooled soil cores per site which were separated by

horizon at the time of sampling. Methods of taking soil cores were given in Jacobson and Band (1987). The number of annual replicates differed across years (Table 4).

As stated above, EM monitoring of the study sites was performed by IITRI (Haradem *et al.*, 1994).

1.3 Results. The sites were similar in biological characteristics, and differed in physical and chemical properties.

Soil pH (Fig. 1) ranged between pH 6 to 7, with most values in the mid-range.

The bulk density of soils was done in 1984. The average density of the mineral horizon and the organic horizon was 1.41 g/cm³ and 0.39 g/cm³ respectively. The average ratio of mineral to organic density was 3.65 (Table 2). These were analyzed by site (Table 2). Pairwise comparisons indicated that organic (ORG) horizon soil at the antenna site differed ($p < 0.05$) from the control (CON) and ground (GND) sites and other comparisons were not significantly different.

Soil suction data (Table 3) provide a rough approximation of the size of water-filled pores at a given moisture content. Although laboratory data for soil moisture retention may not accurately reflect field conditions (Baver *et al.*, 1972). This has been applied to growth of the ciliate *Colpoda* in soil at different moisture levels (Darbyshire, 1975). Growth correlated with sufficient moisture to fill pores the size of the ciliate. In Section 2 (Population Size), data will be presented concerning soil moisture and growth of amoebae. In 1983, when this data was

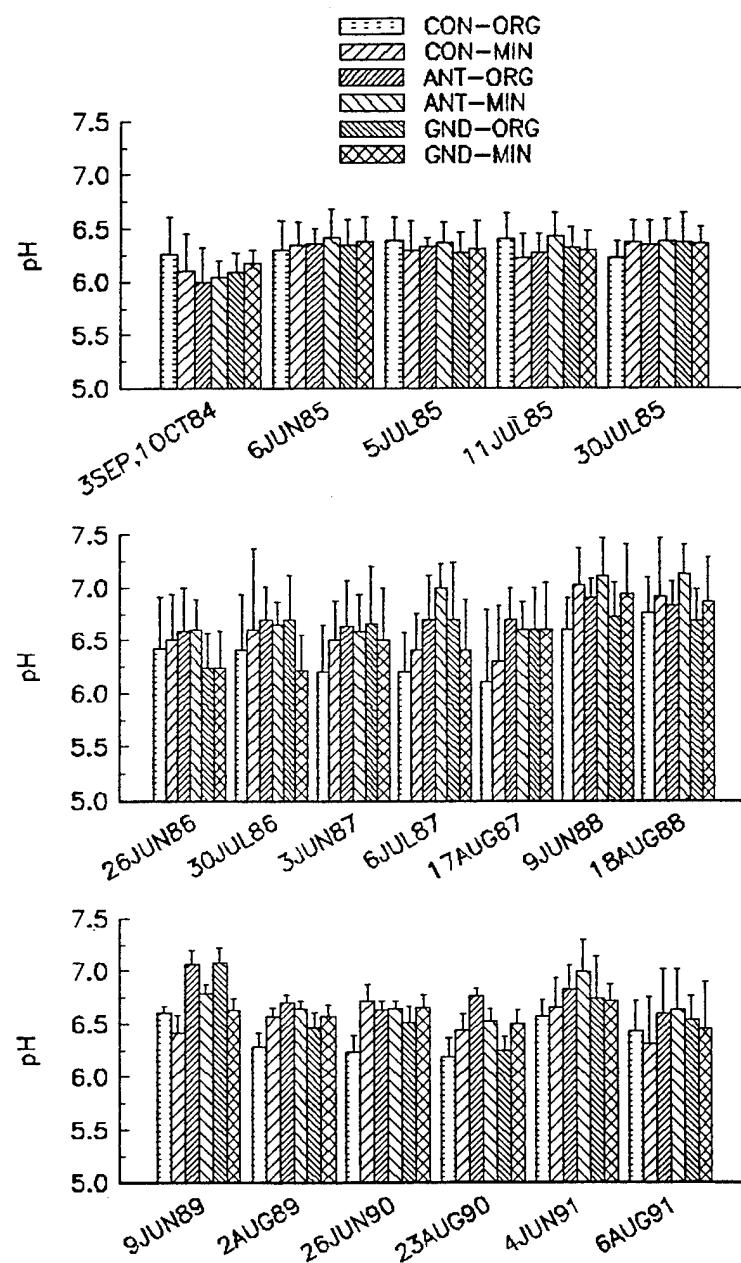


FIGURE 1. Soil pH summary ($\text{mean} \pm \text{SD}$, $n = 10$). Upper graph for pre-operational years, middle graph for intermittent years and lower graph for operational years. Site/horizon abbreviations given in Table 1.

TABLE 2. Mean \pm SD soil bulk density (g/dry wt/cc) of organic and mineral horizons in each study site, and results of ANOVA ($n = 8$ samples/site/horizon).

Site	Horizon	Mean \pm SD	Ratio (MIN/ORG)
Control	Organic	0.39 \pm 0.09	3.69
	Mineral	1.44 \pm 0.06	
Antenna	Organic	0.44 \pm 0.1	3.23
	Mineral	1.42 \pm 0.07	
Ground	Organic	0.32 \pm 0.07	4.25
	Mineral	1.36 \pm 0.09	

One-way ANOVA, Organic horizons:

	D.F.	M.S.	
Between	2	0.02907	
Within	21	0.00767	F=3.79129

Pairwise comparison:

	Uncorrected p value	Bonferroni p value
Control vs. antenna:	0.2726	ns ($p>0.05$)
Control vs. ground:	0.1322	ns ($p>0.05$)
Antenna vs. ground:	0.0159	* ($p<0.05$)

One-way ANOVA, Mineral horizons:

	D.F.	M.S.	
Between	2	0.0139	
Within	21	0.0055	F=2.50547 (ns)

Pairwise comparison:

	Uncorrected p value	Bonferroni p value
Control vs. antenna	0.5992	ns ($p>0.05$)
Control vs. ground	0.0494	ns ($p>0.05$)
Antenna vs. ground	0.1290	ns ($p>0.05$)

ns = not significant

TABLE 3. Soil suction (mean \pm SD, n = 9). Data obtained in 1983 when study sites were not identified. For these measurements soil was taken from a putative antenna which was near the antenna site used for the rest of the study.

0.3 BAR (approximately 10 μm pores fluid filled):

1. Organic horizon: mean = 59% \pm 4.47 H₂O (n=9)
2. Mineral horizon: mean = 21% \pm 0.90 H₂O (n=9)

1.0 BAR (approximately 3 μm pores fluid filled):

1. Organic horizon: mean = 49% \pm 6.04 H₂O (n=9)
2. Mineral horizon: mean = 10% \pm 1.23 H₂O (n=6)

obtained, study sites were not identified; the soil was taken near the antenna study site.

Data on soil moisture (Table 7 and Appendix Tables B-1 to B-10), annual rainfall (Figs. 11, 12) and possible correlations with amoeba growth (Figs. 13 to 16) will be presented in Section 2 of this report.

Average temperature measurements (4 hr intervals) were plotted every third day (Figs. 2, 3, 4). Temperature readings fluctuated between 10 to 20 °C over the season, and was not consistently different between study sites.

Soil chemistry showed variability between sites in past years although general trends in concentrations were seen (Table 4). However, no variation in soil chemistry was evident between sites according to numbers of amoebae, which differed little between sites for a given horizon/date (see Appendix C) for annual summaries of counts and statistical analyses). The chemical analyses were not done in 1992 or 1993.

See Table 1 for a summary of IITRI electromagnetic measurements at the sites over several years (Haradem et al., 1994). The 76 Hz electric field and the magnetic flux density was greater at the antenna site, less at the ground site, and very small at the control site.

2. POPULATION SIZE

The objective was to determine population size of amoebae in soil over the growing season. This is a productivity measure that could be affected by ELF radiation either directly or

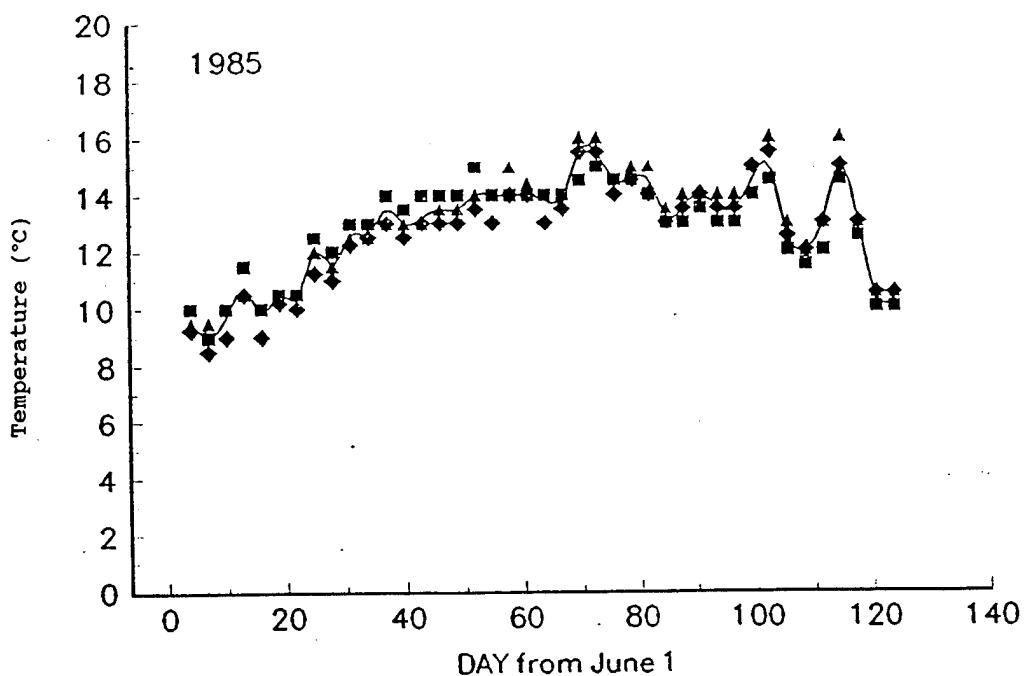
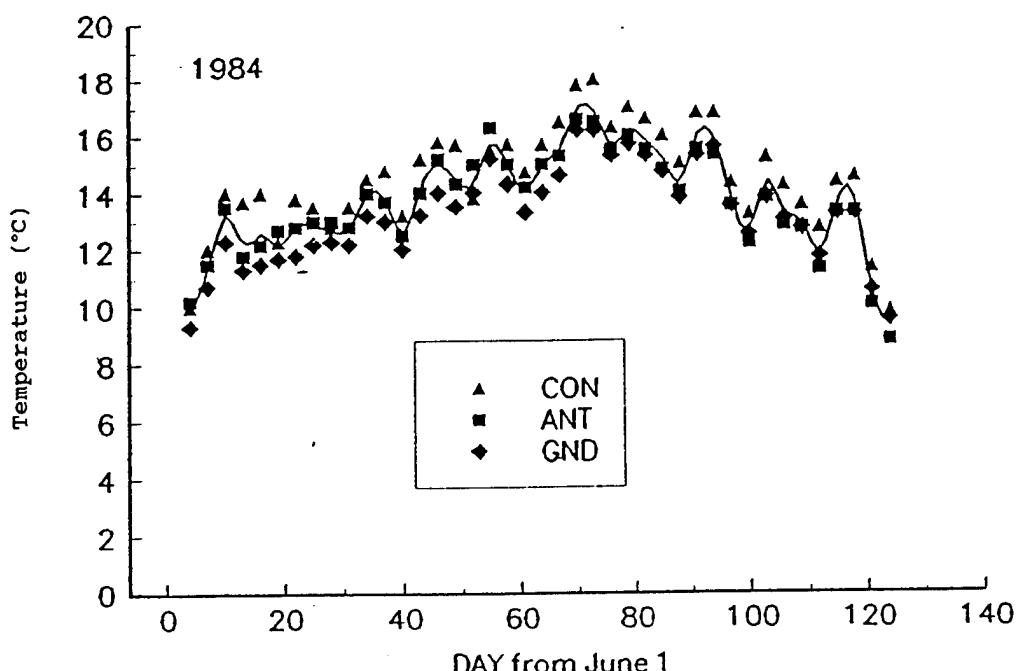


FIGURE 2. Mean soil temperature at three-day intervals, 1984/1985 (pre-operational). Means from measurements recorded every 4 hr.

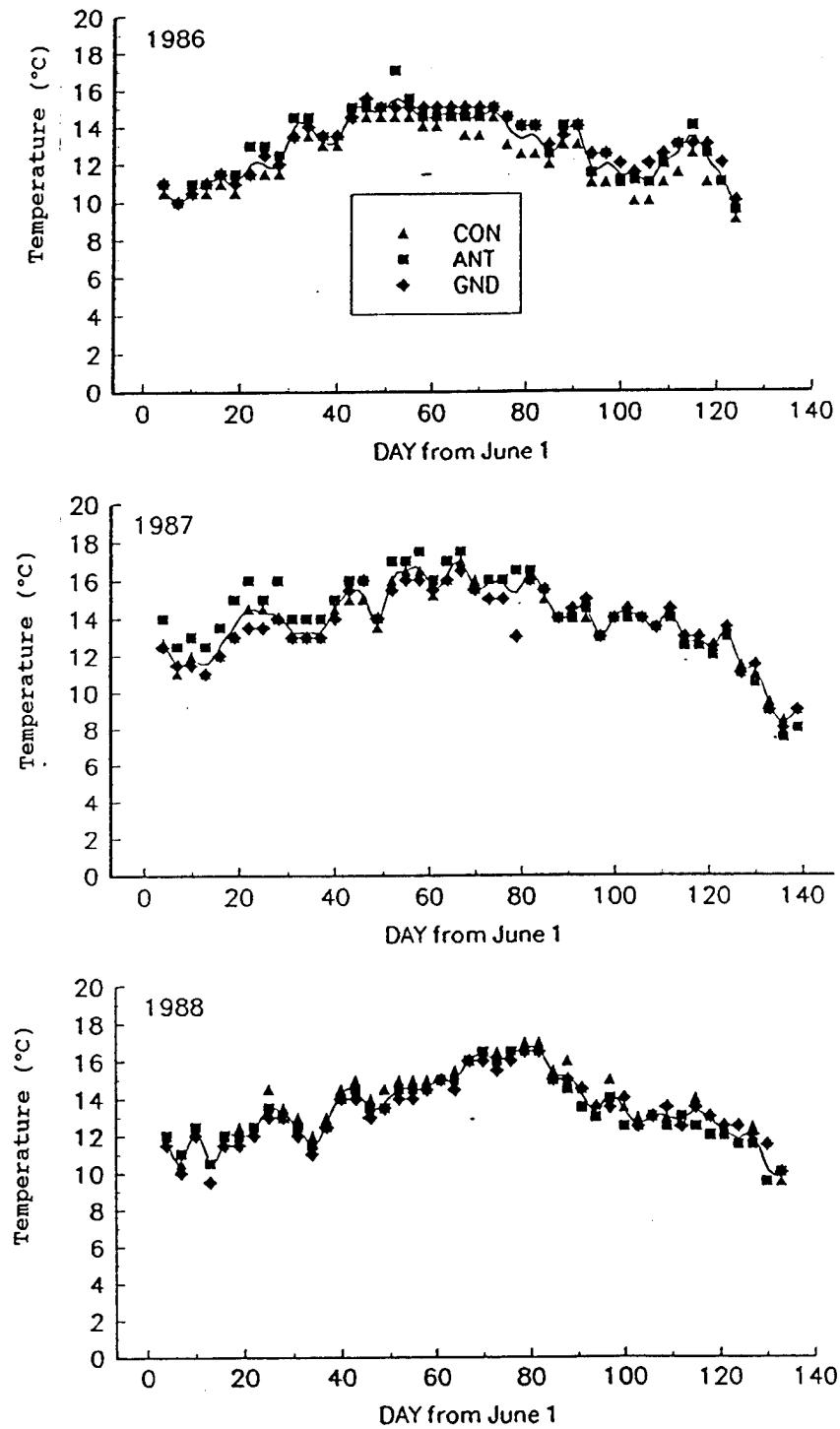


FIGURE 3. Mean soil temperature at three-day intervals, 1986/1987/1988 (intermittent antenna operation). Means from measurements recorded every 4 hr.

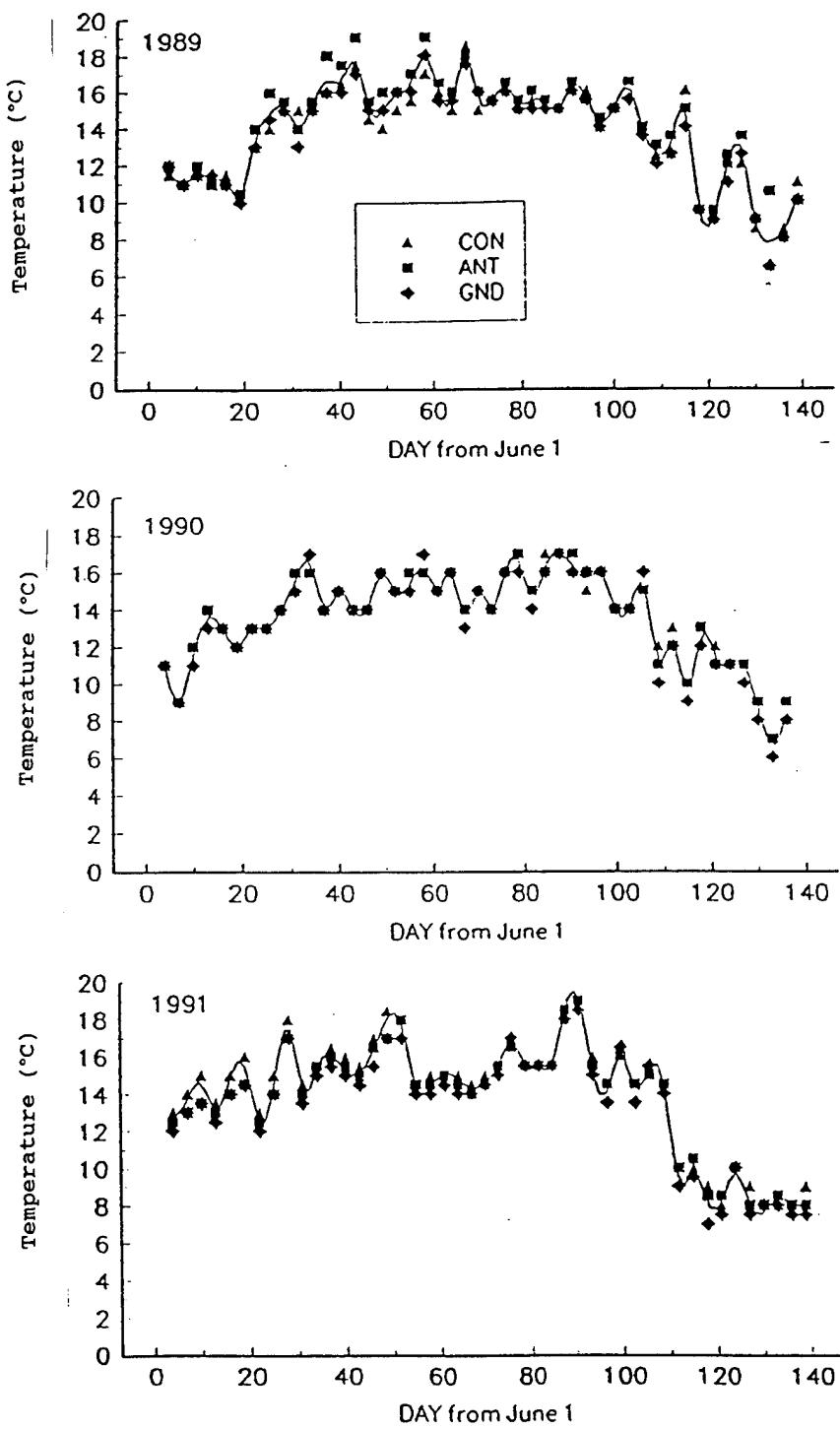


FIGURE 4. Mean soil temperature at three-day intervals, 1989/1990/1991 (operational). Means from measurements recorded every 4 hr.

TABLE 4. Soil chemistry summary^a

Element ^b	Pre-operation (1984-1985)			Transition (1986-1988)			Operation (1989-1991)		
	Control	Antenna	Ground	Control	Antenna	Ground	Control	Antenna	Ground
Organic									
P	32±7	29±7	31±4	41±7	41±8	36±3	36±9	34±5	27±3
K	112±22	115±13	111±23	140±72	124±29	147±57	131±24	140±50	142±34
Ca	2188±548	2264±364	2511±292	2817±1091	2535±255	3019±790	2480±685	2660±614	2744±842
Mg	156±33	151±15	162±30	182±83	169±20	208±55	176±47	200±78	227±66
NO ₃	13±4	15±7	20±10	11±10	13±13	11±13	5±2	5±2	5±2
Organic N	13±7	12±3	12±3	6±1	6±1	6±1	7±2	7±1	7±1
Mineral									
P	60±14	53±8	37±20	60±28	35±25	23±4	49±25	33±26	23±3
K	36±10	24±5	28±9	35±2	35±4	33±2	40±4	39±5	40±3
Ca	583±74	478±175	595±170	728±110	785±95	826±87	742±103	865±61	824±164
Mg	53±9	32±21	45±21	72±21	82±11	79±17	74±10	89±13	87±15
NO ₃	2±2	3±2	1±2	5±2	9±9	9±11	3±2	3±2	3±2
%Organic N	1±0	1±0	1±0	1±0	1±0	2±0	2±0	2±0	2±0

^aSoil chemistry was assessed once in 1984 (n/site = organic, 3; mineral, 4) and 1985 (n/site = 4) and twice in other years (n/site for each sampling = 2).

^bData expressed as ppm except for organic N, which is in percent.

through changes in the number or type of microbes used as food for the amoebae. The ratio of vegetative to dormant amoebae was also determined as an estimate of biological activity of the amoebae in soil. Determinations of population size were made during each growing season (June to October) from 1984 to 1991 and at a reduced level in 1992. Few differences were noted between study sites before or after the antenna began operation. Soil amoeba populations at all sites exhibited a significant, annual, increase in population size from June through July or August followed by a decrease in the autumn. The annual, maximum total population size was in part related to soil moisture.

2.1 Methods.

2.1.1. *Sampling.* Eight random soil samples were taken from each site with a 2 cm diameter tube sampler, with 4 to 6 cm of soil taken from each horizon in a single sample (Jacobson and Band, 1987). Each sample contained the ORG and MIN horizons, which were separated at the time of sampling prior to processing in the laboratory. Samples were processed in the lab within 3 hr of sampling at sites.

Each soil horizon (ORG and MIN) per site was subdivided into two, 1 g (wet weight) portions. A third portion of the soil sample was used to determine moisture content as described in section 1.2, above. The two, 1 g portions were suspended in low salt saline (LSS): 50 mM NaCl, 4.6 mM MgSO₄, 0.36 mM CaCl₂ (Band and Mohrlok, 1969). One portion was processed for counting directly while the other was treated with 1% HCl for 24 hr to kill vegetative amoebae but not encysted amoebae. The 1% HCl was

not a real 1% (v/v) solution but rather a hundredfold dilution of concentrated HCl. After 24 hr, the HCl-treated portion was washed by centrifugation (approximately 800 g) three times in LSS to remove the HCl and then processed for counting. The soil portion used without HCl treatment (total counts) was also washed three times by centrifugation to assure that both portions were equally dispersed for counting. After washing, the samples were suspended in 5 ml of LSS for counting.

2.1.2 *Counting*. An established soil dilution counting procedure was used (Singh, 1946; as modified by Darbyshire et al., 1974). The method utilized Fisher's table of densities of organisms estimated by the dilution method (Fisher and Yates, 1963). Each sample was subdivided into 8 replicates, which were twofold diluted for a series of 12 dilutions. This still represented a single sample. It was thus appropriate to do statistical analyses on replicate samples per site/horizon/date.

Prior to 1989 the amoebae were enriched for soil dilution counting by being fed with living *E. coli* (K12) at a concentration of 1 mg/ml. A similar approach was used for growth experiments described later. Since 1989, lyophilized *E. coli* (Sigma Chem. Co., EC-11303), sterilized by ⁶⁰Co irradiation (282,000 R), was used as the food source at a concentration of 1 mg/ml. Differences between total amoebae and cysts were used to determine total vegetative amoebae.

2.1.3 *Sample number*. A preliminary soil count, with ten samples from each horizon at the three sites, was made in 1983 (Table 5). The results indicated a coefficient of variation that

TABLE 5. Estimates of total amoeba densities per site and horizon, September 7 and October 16, 1983 (mean \pm SD log no/g soil, n = 10).

SITE	HORIZON	DATE	MEAN \pm SD	COEF. OF VARIATION
Control	Organic	9/7	3.79 \pm 0.21	5.5%
		10/16	3.78 \pm 0.33	8.7%
	Mineral	9/7	2.89 \pm 0.28	9.7%
		10/16	3.14 \pm 0.18	5.7%
Antenna	Organic	9/7	3.32 \pm 0.14	4.2%
		10/16	3.44 \pm 0.14	4.1%
	Mineral	9/7	3.05 \pm 0.21	6.9%
		10/16	3.10 \pm 0.39	12.6%
Ground	Organic	9/7	3.50 \pm 0.21	6.0%
		10/16	3.68 \pm 0.25	6.8%
	Mineral	9/7	2.98 \pm 0.33	11.1%
		10/16	3.11 \pm 0.10	3.2%

was <10% of the mean or less for a given horizon and date, with two exceptions (i.e., 11.1 and 12.6%). Thus from a 90% power curve, significant differences could be detected at 1.4 x standard deviation (SD) for a sample size of 10 and 1.5 to 1.6 x SD for a sample size of 8. Thus sample sizes of 8 and 10 were almost equally powerful so that 8 random samples were taken from each horizon at the three sample sites on a sampling date.

Other published studies with this method used fewer counts however the authors were not testing site differences.

Darbyshire et al. (1974) and Elliott et al. (1980) used single samples; Bryant et al. (1982) did duplicate counts per sample.

2.1.4 **STATISTICS.** One-way analysis of variance was used to detect site differences (control, antenna and ground sites for each horizon) in total amoeba and cyst counts (Appendix Table C-3). Data were transformed by logarithm to achieve greater conformance to the normality assumptions for the residuals of the analysis.

The Before-After-Control-Impact (BACI) analysis suggested in Steward-Oaten et al. (1986) starts with the model with measurements on a variable taken at specific sites Control and Impact at specific times Before and After an intervention that is thought to possibly effect the variable at the Impact site and not at the Control site. The authors discuss an analysis which in its simplest form is the t-test that compares the average of differences Impact-Control at times before the intervention with the average of differences Impact-Control at times after the intervention. In this simple model, the differences are

considered to be independent random variables and these random variables are assumed to have a constant mean and variance before the intervention and a possibly changed mean but the same variance after the intervention. The t-test tests the hypothesis of no change in the mean from before the intervention to after the intervention against the alternative hypothesis that there is a change. The authors mention common diagnostic techniques to check assumptions and transformations and variations on the data analysis to meet departures from model assumptions.

In the following we analyzed several response variables using the BACI method. For the BACI analysis of log "maximum amoeba count" and log "maximum cyst count" in the organic horizon, the Before period consisted of the years 1984-1988 and the After period consisted of the years 1989-1992. For the BACI analyses of log "maximum amoeba count" and log "maximum cyst count" in the mineral horizon, the Before period consisted of the years 1984-1988 and the After period consisted of the years 1989-1991. The BACI analysis was also used in Section 5.

Direct counts of amoebae in soil, as is done with freshwater organisms (e.g., Wright and Coffin, 1984) was not possible (Heal, 1970; Heal, 1971).

2.2 Results.

2.2.1 *Counts.* Few significant differences were noted in total counts for a given soil horizon between the research sites (Figs. 5 to 8, Table 6 and Appendix Table C-3), while cyst counts exhibited more significant differences (Table 6 and Appendix Table C-3). These will be described below (Section 2.2.3).

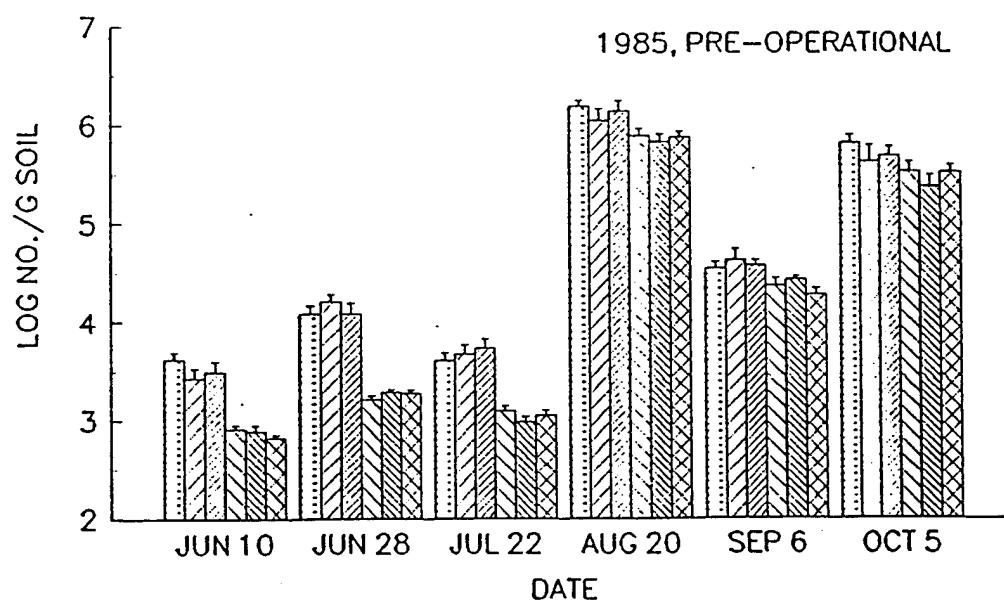
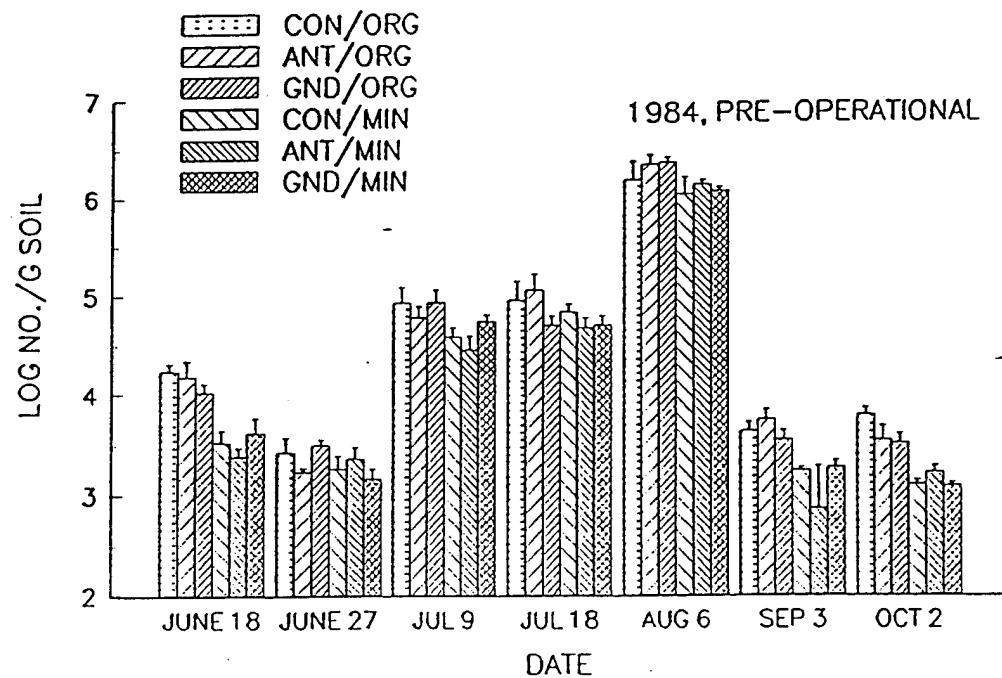


FIGURE 5. Total amoeba counts, 1984/1985, pre-operational (mean \pm SE, n = 8).

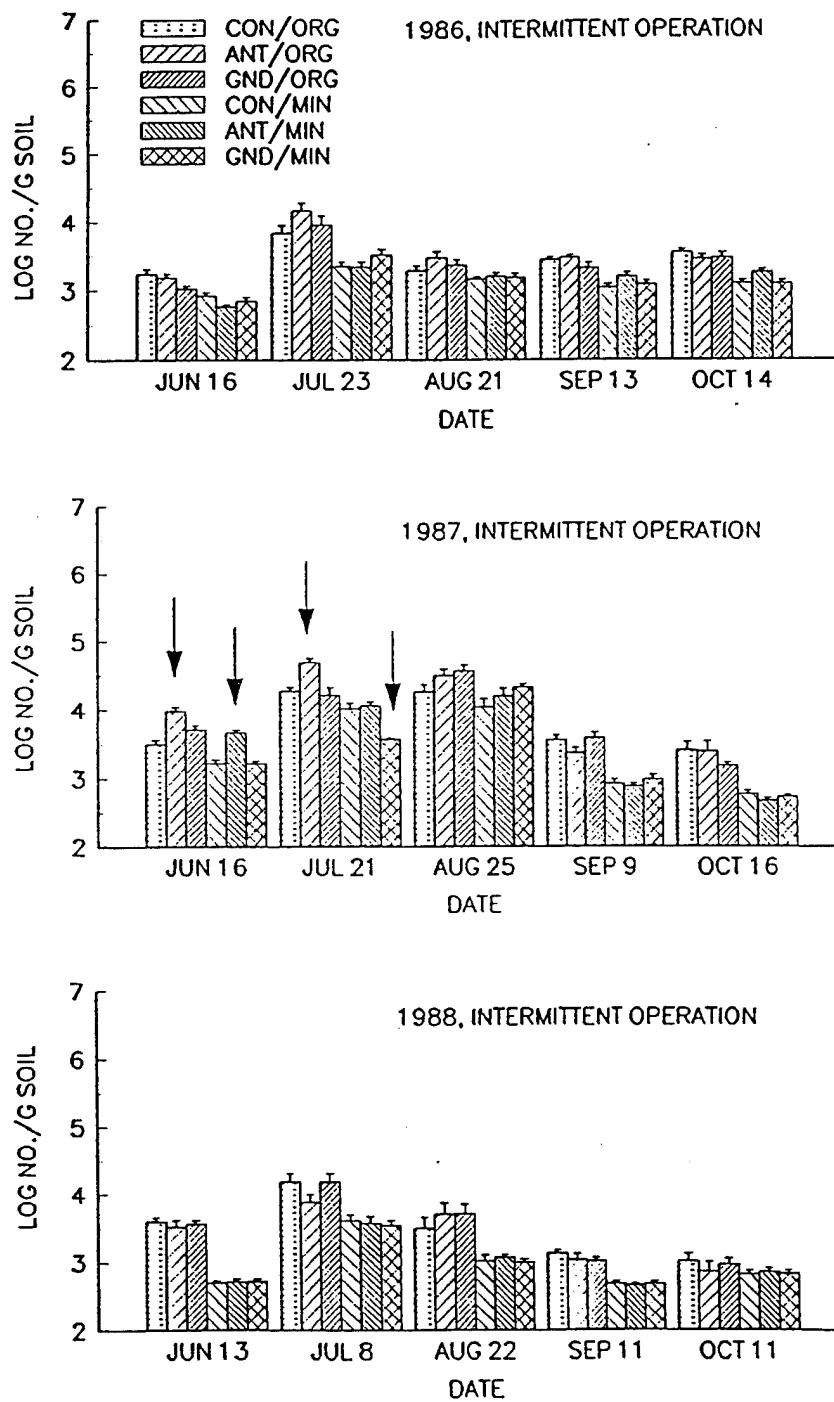


FIGURE 6. Total amoeba counts, 1986/1987/1988, intermittent operation (mean \pm SE, n = 8). Arrows indicate significant differences between sites for a given date and horizon, see Appendix C for data and analysis.

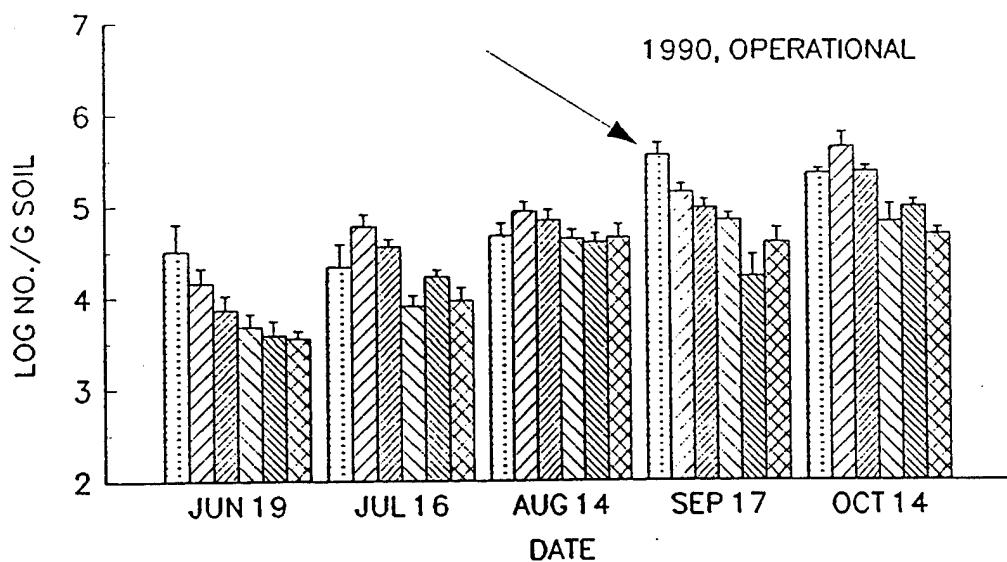
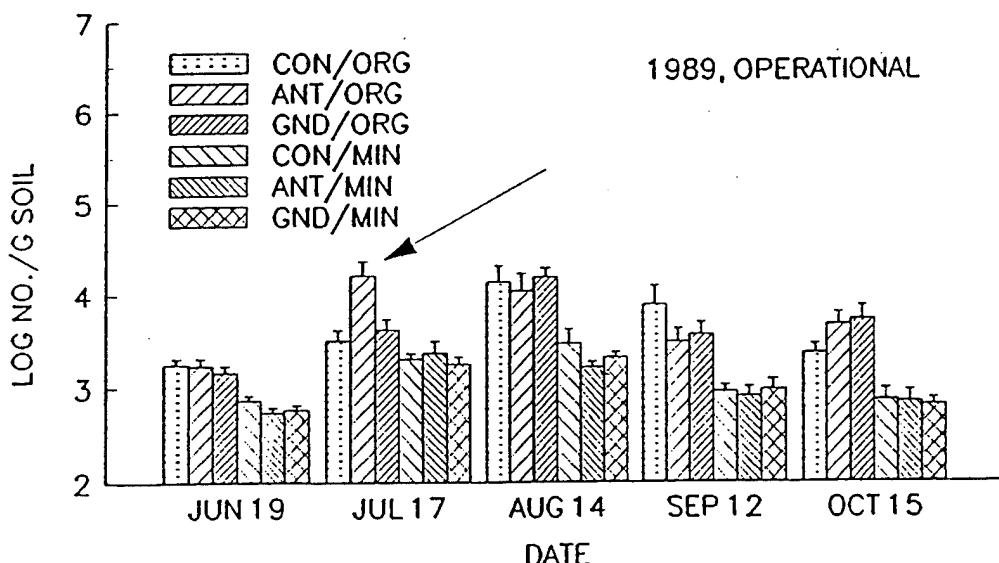


FIGURE 7. Total amoeba counts, 1989/1990, operational (mean \pm SE). See Fig. 6 for explanation of arrows.

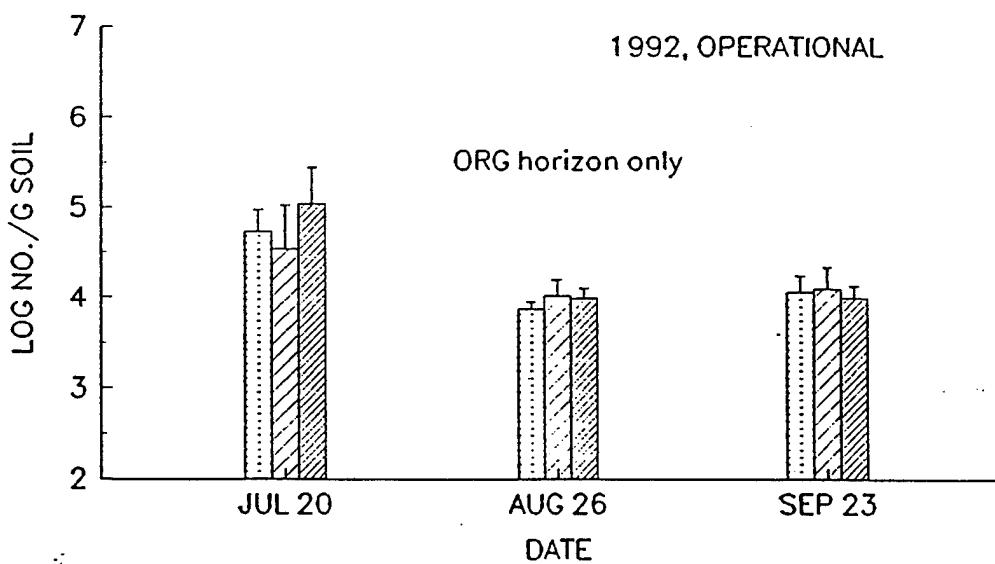
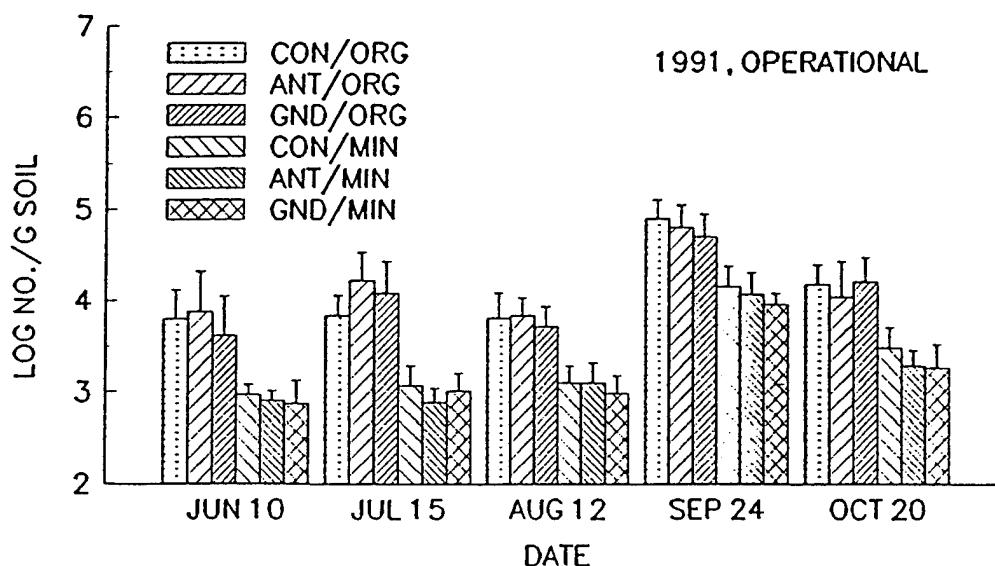


FIGURE 8. Total amoeba counts, 1991/1992, operational (mean \pm SE).

TABLE 6. Number of monthly sample sets where a site was significantly different for total counts or cyst counts. Data taken from Appendix Table C-3.^a

Year	Total Counts				Cysts			
	Organic horizon		Mineral horizon		Organic horizon		Mineral horizon	
	Differ n/yr. ^b	Differ n/yr.	Differ n/yr.	Differ n/yr.	Differ n/yr.	Differ n/yr.	Differ n/yr.	Differ n/yr.
(Pre-operational)								
1984	0	7	0	7	4	7	4	7
1985	0	6	0	6	2	6	4	6
1986	0	5	0	5	1	5	1	5
1987	2	5	2	5	0	5	1	5
1988	0	5	0	0	1	5	0	5
(Operational)								
1989	1	5	0	5	2	5	1	5
1990	1	5	0	5	3	5	0	5
1991	0	5	0	5	2	5	4	5
1992	0	3	not done		1	3	not done	

^aIllustrated in Figs. 6, and 7 for total counts.

^bNumber of monthly sampling dates when one site was significantly different from the other sites (Differ), compared to the total number of monthly samples for the year (n/yr.)

Count data is also presented in Appendix Tables C-1 and C-2.

2.2.2 Seasonal cycle. Soil amoeba populations annually increased from the start of the growing season to an observed maximum, which in August of 1984 and 1985 was in excess of a million amoebae per gram of soil. Then the amoeba populations decreased in the autumn to a few thousand/gram soil (Figs. 5, 10). Subsequent years did not exhibit populations of this magnitude, but they still exhibited a similar cyclic pattern of seasonal growth. Total count fluctuations in the mineral horizon paralleled those in the organic horizon at a lesser population density (Figs. 5 to 8, 10). Note that in 1990 the population decreased sometime after the Oct. 14 count (Figs. 7, 10).

2.2.3 Annual populations. The time of population maxima differed across years (Figs. 9, 10). The drought years from 1986 to 1989 (Figs. 11, 12) coincided with small, annual population sizes of soil amoebae (Figs. 9, 10). No correlation was observed between soil moisture and population size when all data for a given year was compared. Comparisons of moisture measurements of the organic horizon, done on the same sample from which annual population maxima were observed, revealed a correlation between moisture and population size (Fig. 13): correlation coefficient (r) = 0.7047, p value (two-tailed) 0.0001. This is not a strong correlation; note that there is a deviation from linearity around 35% moisture. For individual sites (Figs. 14, 15, 16) the correlation coefficients were: control, r = 0.7734, p = 0.0145; antenna, r = 0.5950, p = 0.0149; ground, r = 0.4805, p = 0.0384. Annual population maxima in the mineral horizon did not correlate

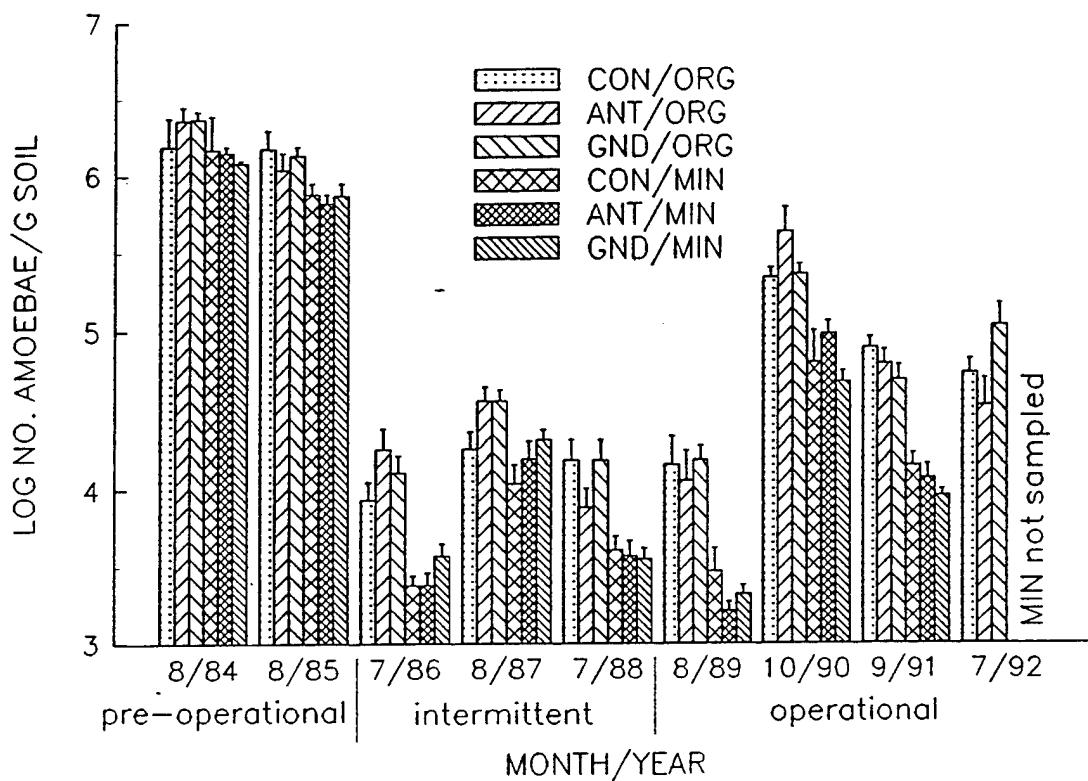


FIGURE 9. Maximum number of total amoebae (means \pm SD/g soil) observed per year, with the sampling date given on the horizontal axis. Abbreviations refer to the study sites (CON, control site; ANT, antenna site; GND, ground site) and to the soil horizons (ORG, upper organic; MIN, lower mineral).

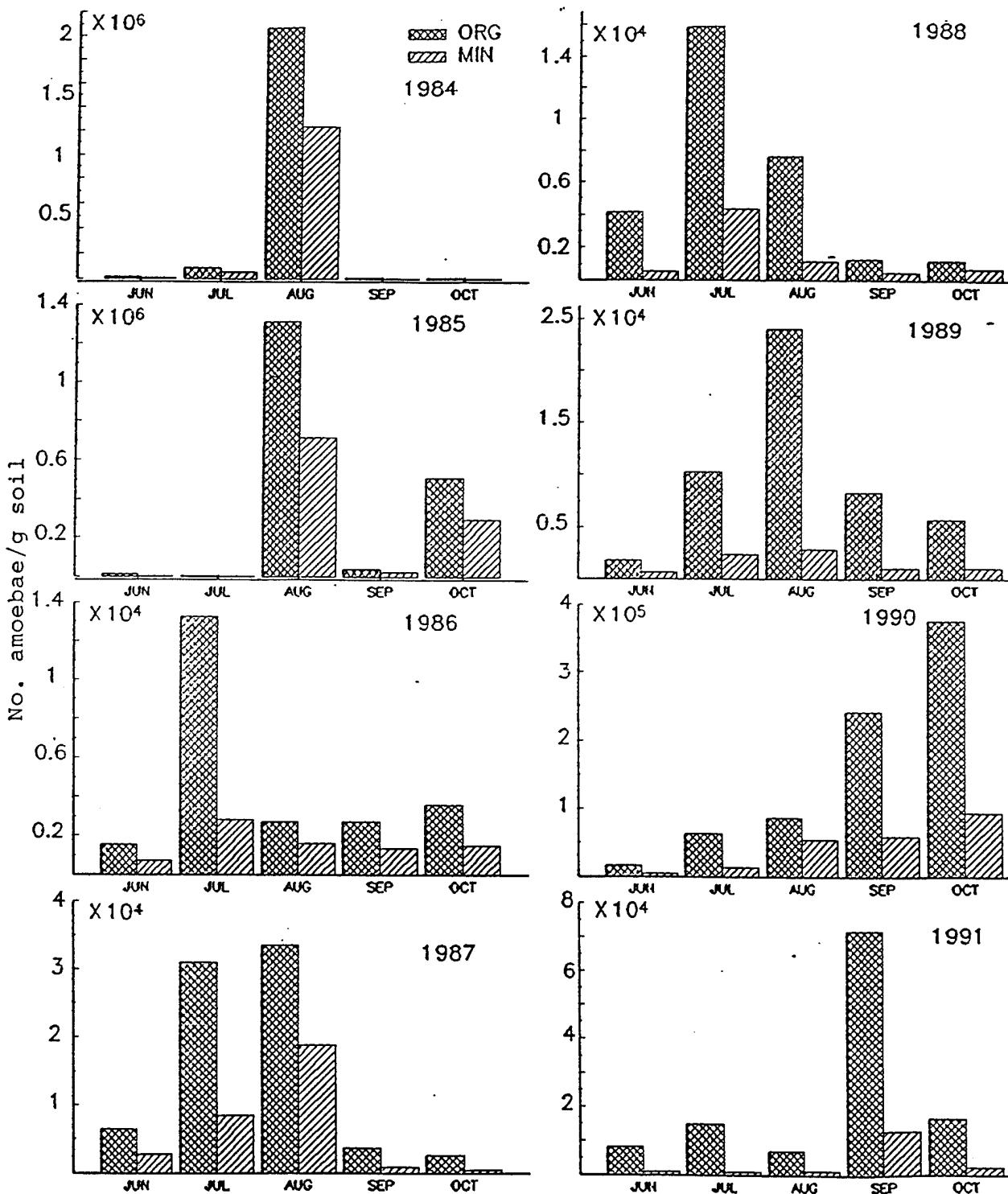


FIGURE 10. Seasonal soil amoeba population peaks. Arithmetic data from pooled means from all study sites presented by soil horizon. See text Figs. 5 to 8 for data plotted (log mean \pm SE) by individual site/horizon. The limited data for 1992 (Fig. 8) were not plotted here.

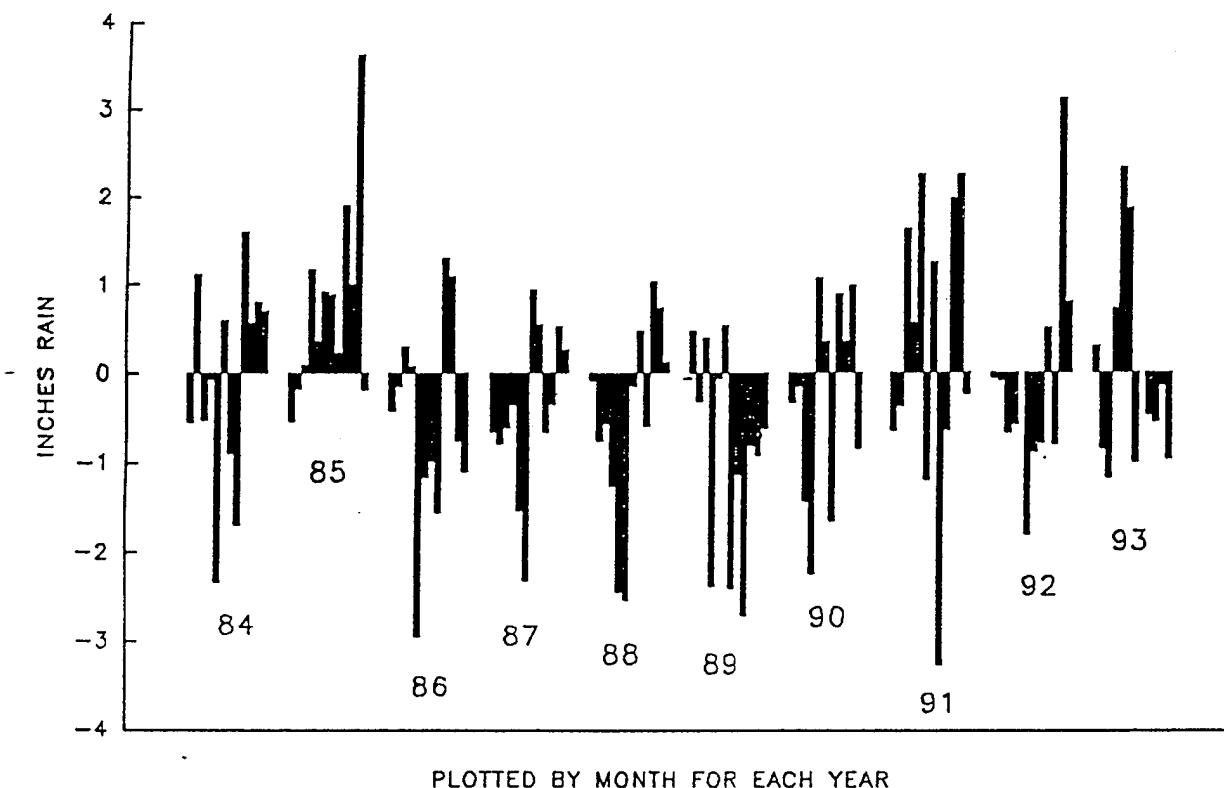


FIGURE 11. Monthly departure from normal (average) rainfall from 1984 to 1993. Data from the National Oceanic and Atmospheric Administration "Climatological Data" for Iron Mountain, Michigan, located approximately 10 to 20 miles south of the study sites. Monthly averages were calculated from 1951-1980, except for 1993 which was the average from 1961 to 1990.

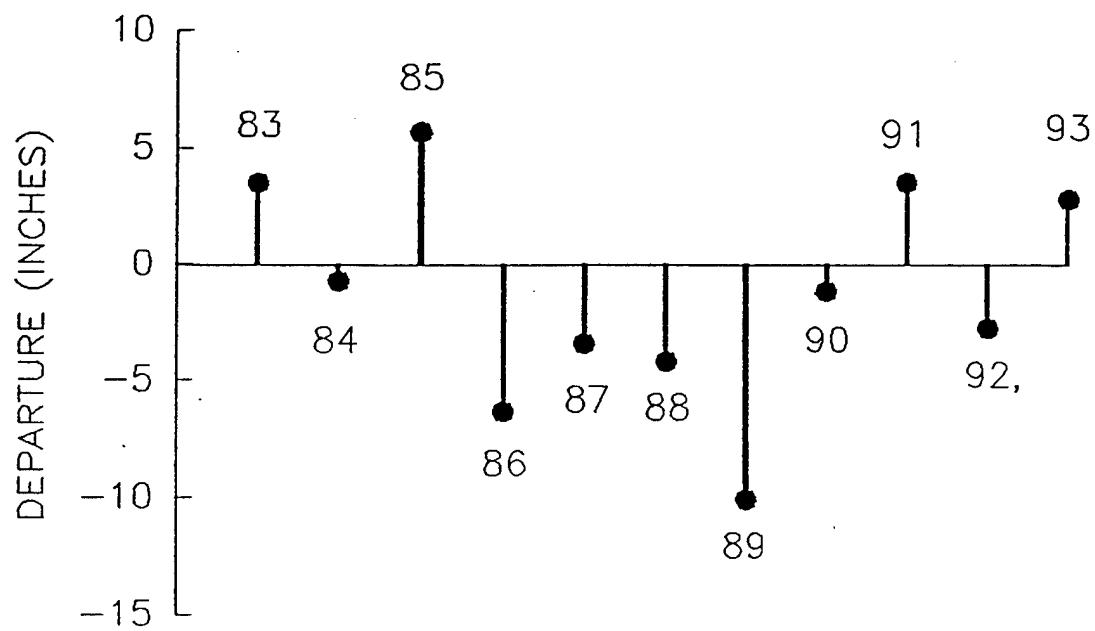


FIGURE 12. Annual departure from normal (average) rainfall from the same data used to plot Fig. 11.

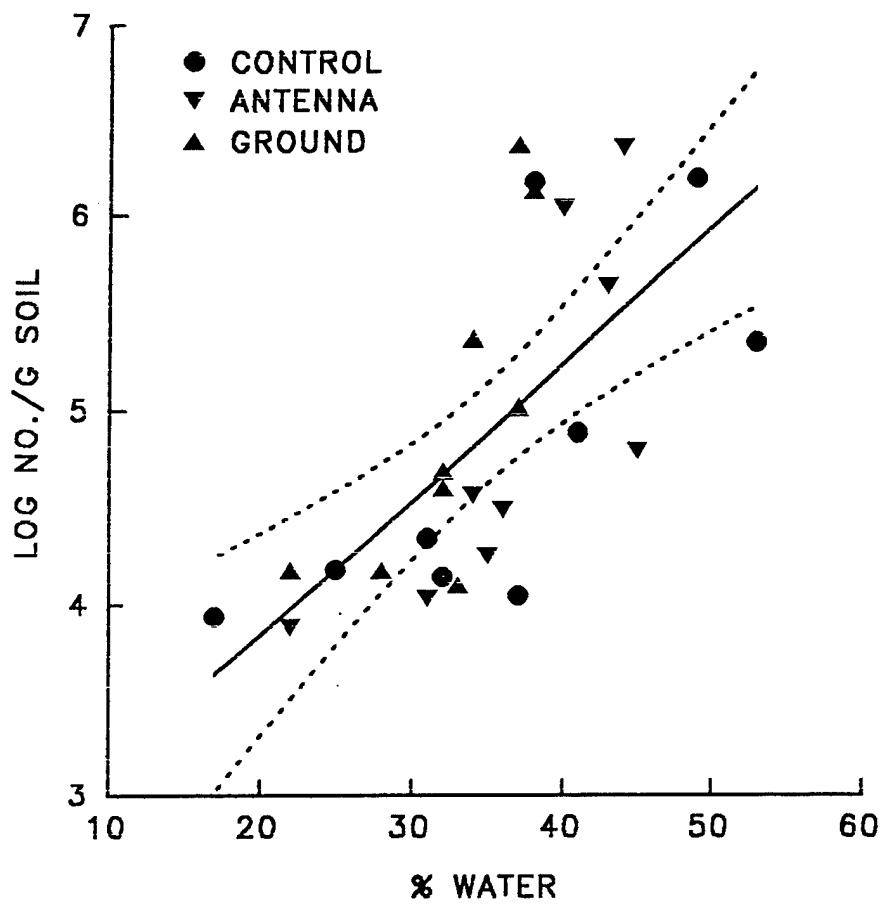


FIGURE 13. Observed maximum, mean, annual amoeba population densities in the organic horizon plotted against mean soil moisture content, 1984 to 1992. Data show for the three sites.

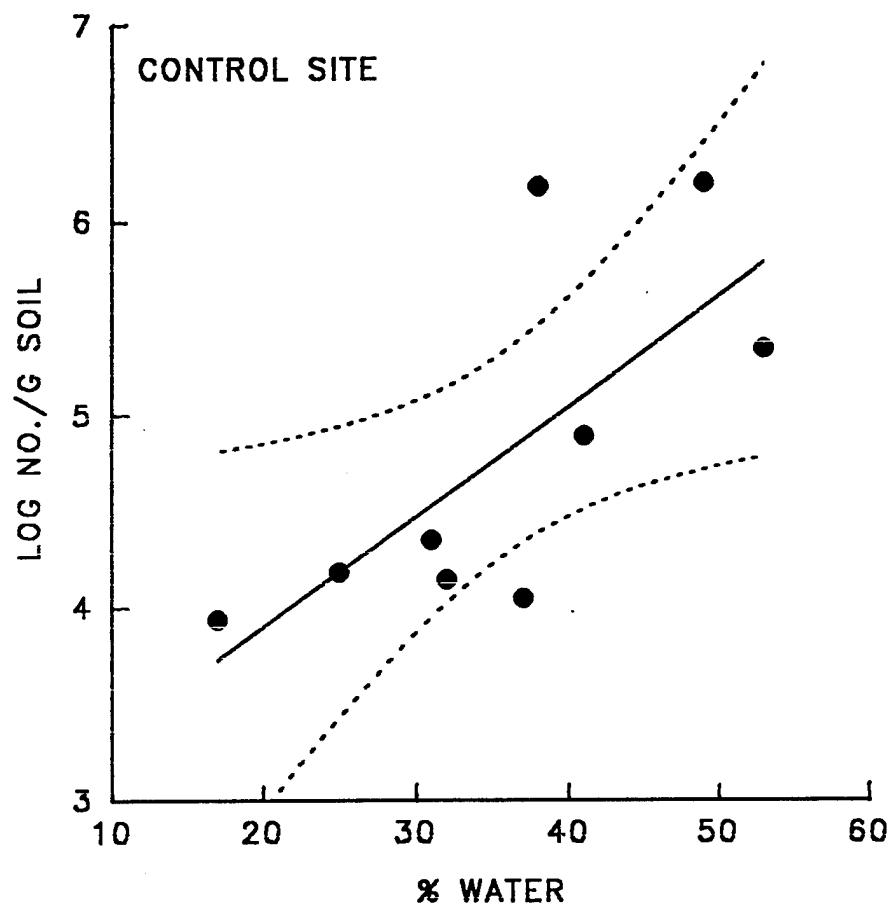


FIGURE 14. Observed maximum, mean, annual amoeba population densities in the organic horizon plotted against mean soil moisture content, 1984 to 1992. Data show for the control site.

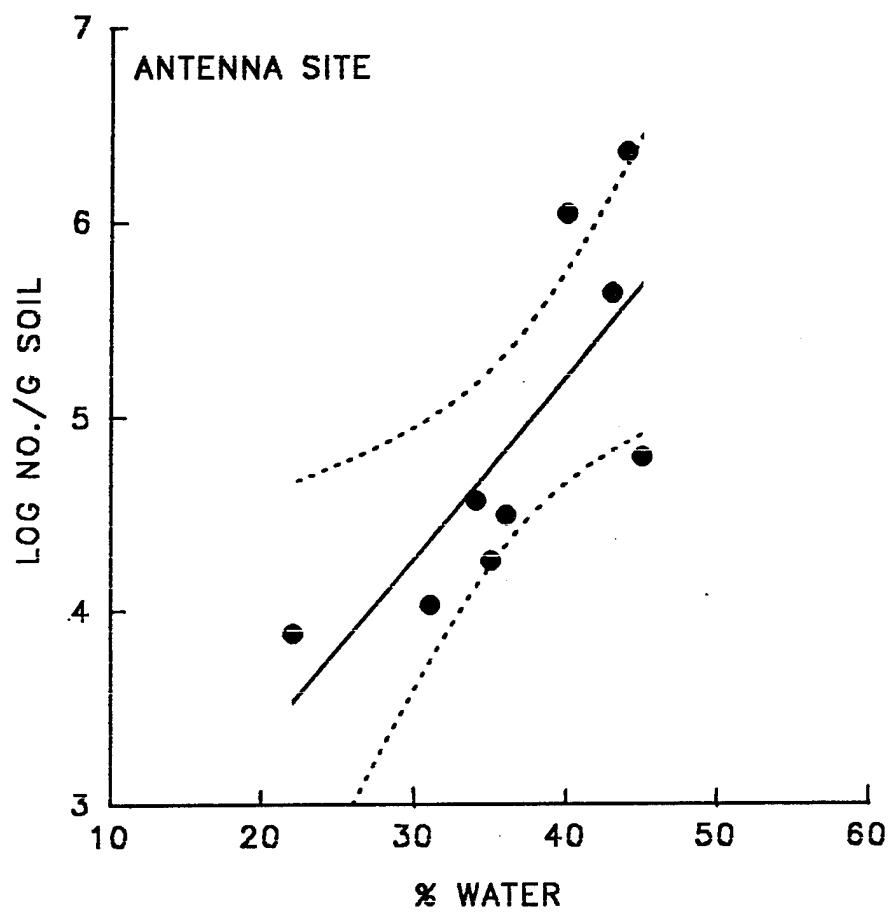


FIGURE 15. Observed maximum, mean, annual amoeba population densities in the organic horizon plotted against mean soil moisture content, 1984 to 1992. Data show for the antenna site.

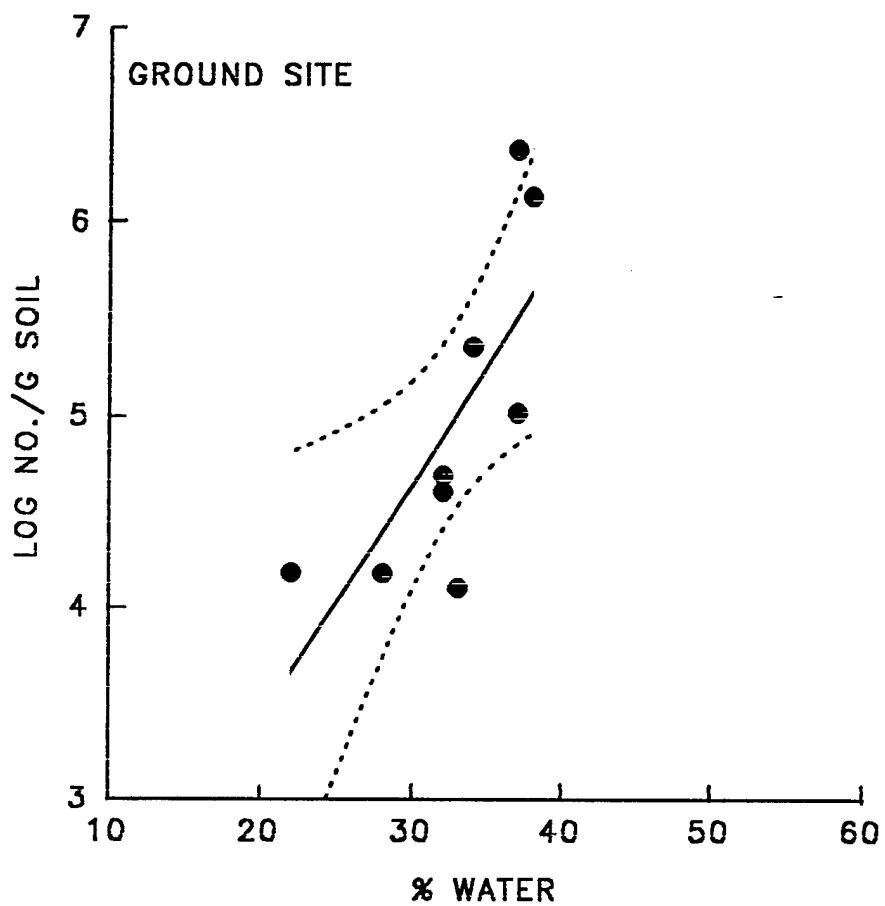


FIGURE 16. Observed maximum, mean, annual amoeba population densities in the organic horizon plotted against mean soil moisture content, 1984 to 1992. Data show for the ground site.

with soil moisture: correlation coefficient (r) = -0.1917, p value (two-tailed) 0.3696.

Few differences were noted in total counts for a given soil horizon between the antenna, ground and control sites (Table 6; Appendix Table C-3). In 1987 the June and July counts were greater at the antenna site for the organic horizon and the June, mineral horizon while the July, mineral horizon at the ground site was less than the other sites (Fig. 6); in 1989 the July count in the organic horizon was greater at the antenna site; in 1990 the September count in the organic horizon was greater at the control site. Otherwise no significant differences in total counts of amoebae were noted between sites for a given horizon and date from 1984 to 1992. Site differences were not observed for annual, maximum population counts (Fig. 10). Vegetative amoebae formed a significant component of each year's observed population maximum (Figs. 17 to 20).

Cyst counts exhibited more significant site differences (Table 6; Appendix Table C-3) possibly due to the vegetative amoeba responding to the lack of food (Band, 1963). Encystation in most amoebae is in response to lack of food and is reversible; it is not related to sexual reproduction. Thus the number of cysts in proportion to vegetative amoebae reflects local availability of food (Band, 1963).

2.2.4 Interaction with other microbes. Approximately 300 isolates of actinomycetes and fungi were enriched from the study sites. Most were eaten or ignored by the amoebae and a few were toxic. One of the actinomycetes that were toxic to the amoebae

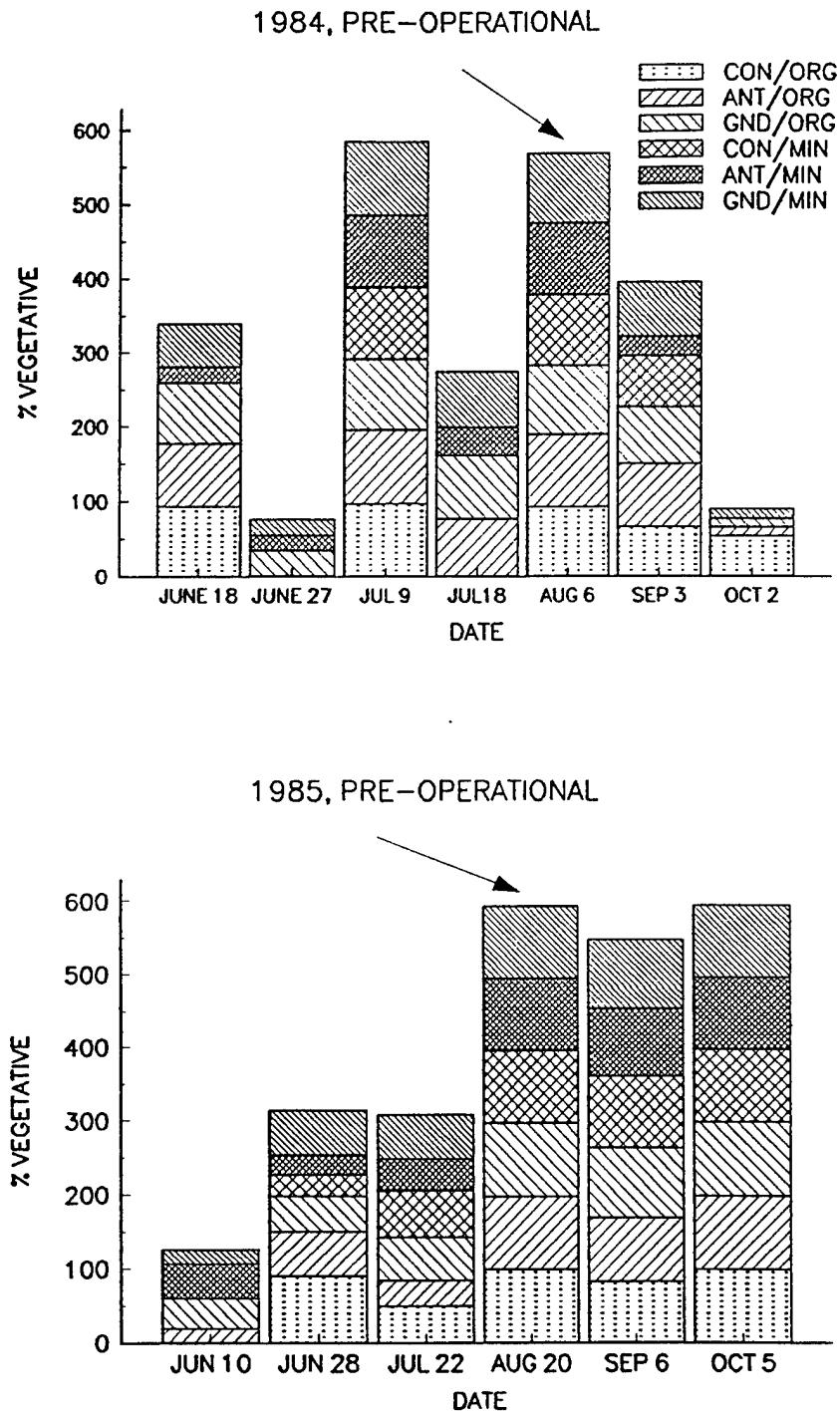


FIGURE 17. Percent vegetative amoebae for each sampling date plotted from total amoeba counts and cyst counts given in Appendix C. Site abbreviations given in Glossary and Fig. 9. Arrow indicates sample with the maximum, total population for each year. Data given for pre-operational years 1984 and 1985.

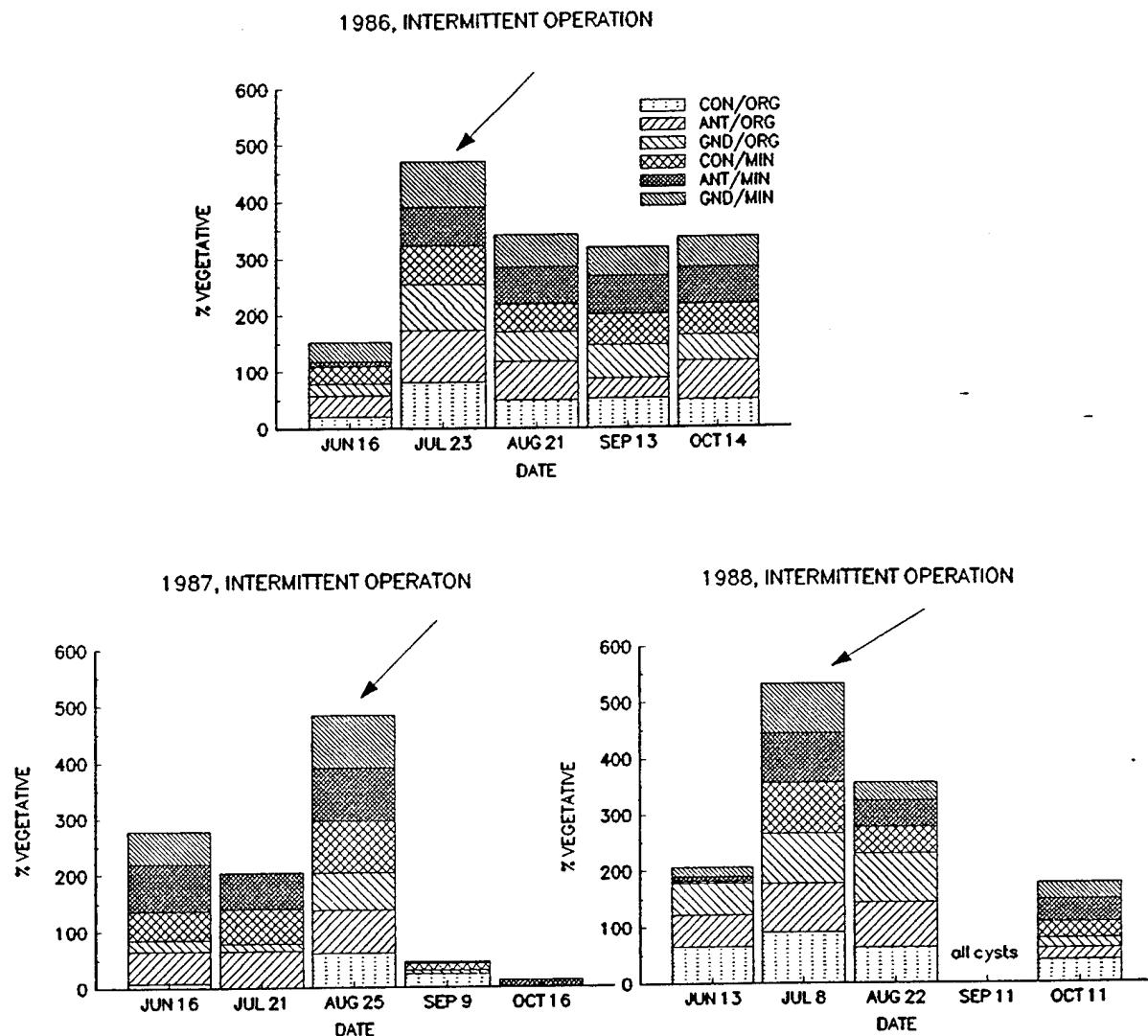
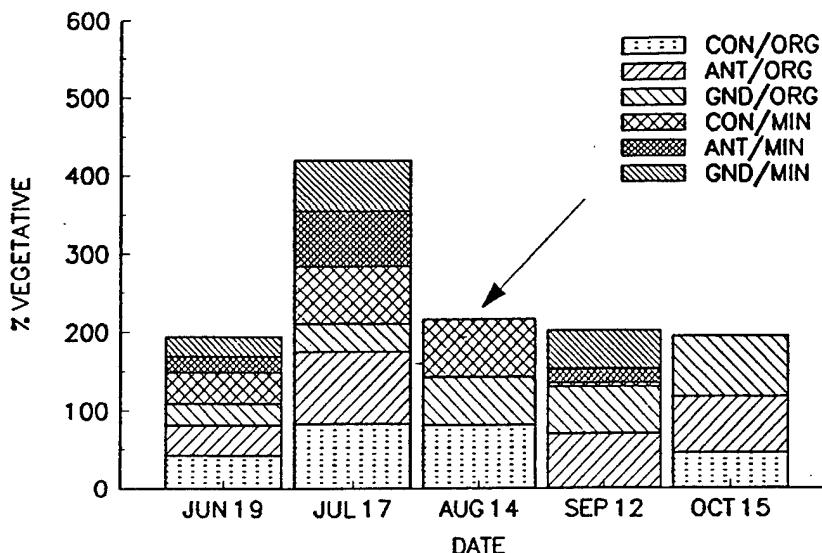


FIGURE 18. Percent vegetative amoebae for each sampling date plotted from total amoeba counts and cyst counts given in Appendix C. See Fig. 17 for details. Data given for intermittent operation years 1986, 1987 and 1988.

1989, OPERATIONAL



1990, OPERATIONAL

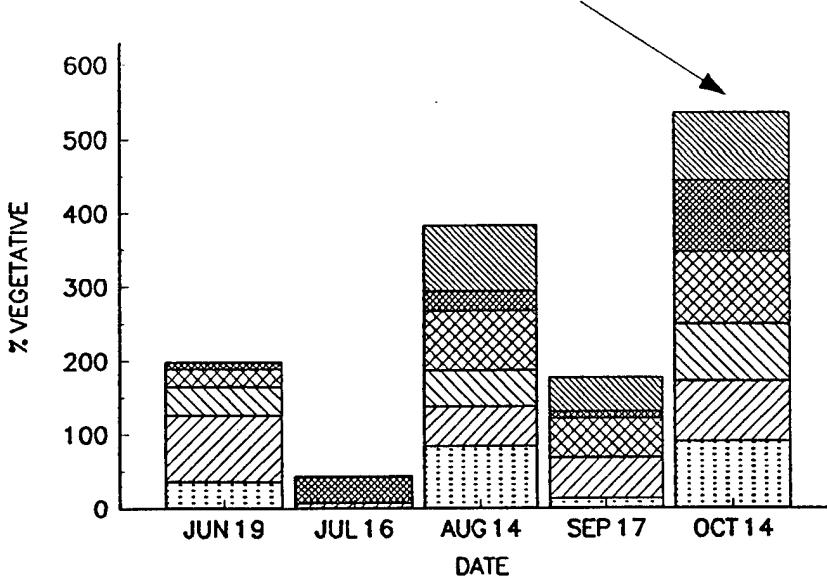
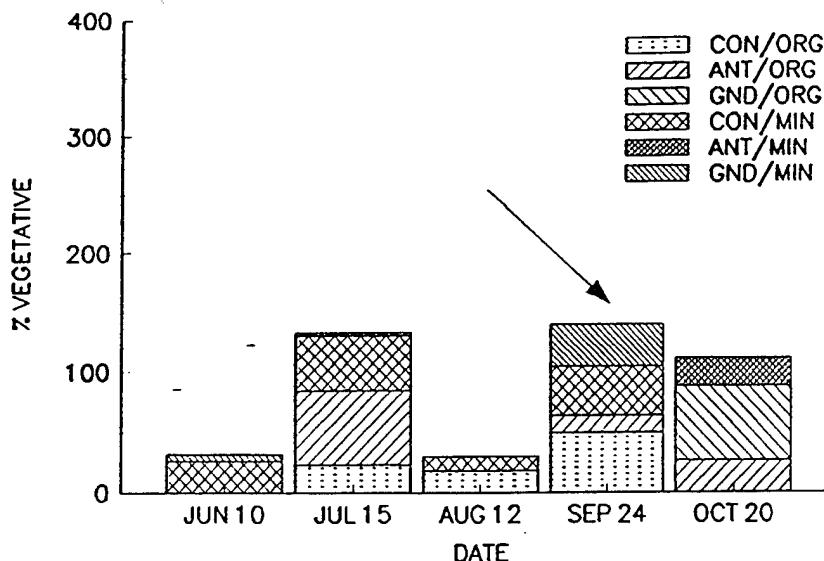


FIGURE 19. Percent vegetative amoebae for each sampling date plotted from total amoeba counts and cyst counts given in Appendix C. See Fig. 17 for details. Data given for operational years 1989 and 1990.

1991, OPERATIONAL



1992, OPERATIONAL

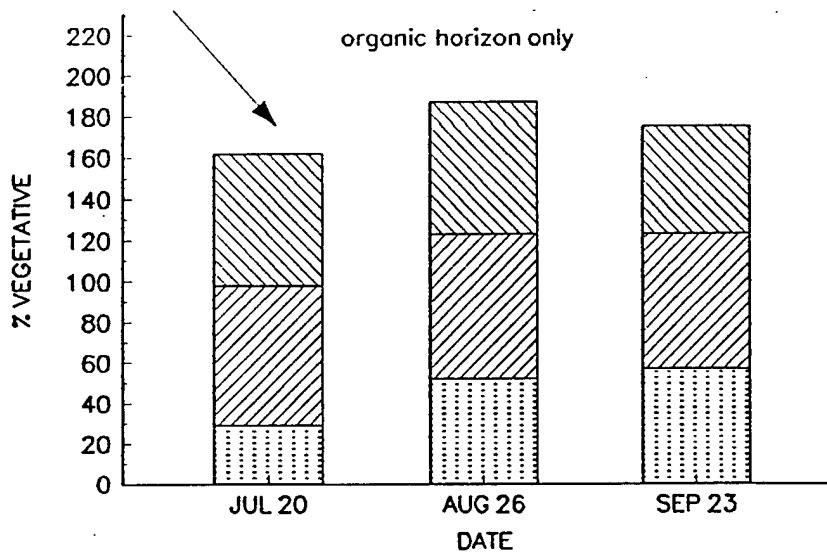


FIGURE 20. Percent vegetative amoebae for each sampling date plotted from total amoeba counts and cyst counts given in Appendix C. See Fig. 17 for details. Data given for operational years 1991 and 1992.

was identified by the American Type Culture Collection, which identified it to genus, i.e. *Streptomyces*, as a strain of the gray color series with spiral aerial mycelia. Strains in this group are commonly used to produce antibiotics and toxins commercially, and this has resulted in a very complex taxonomy. Thus a detailed taxonomic classification of this isolate would not be practical. However, this does illustrate a biological basis for proposing that other microbes in soil may be responsible for controlling amoeba populations.

Two attempts were made to estimate bacterial population sizes in soil to see if these also cycled annually. First was an attempt to develop a direct bacterial count method. Indirect biochemical methods of estimating biological content of soil include an estimate of the total soil biota, rather than a unit of bacteria. A combination of a method for estimating bacteria in aquatic environments (Hobbie et al., 1977) and isolation of fungi from soil (Hanssen et al., 1974) was attempted. The method involved blending a small quantity of soil (0.5 g) in 1 l of 2% formaldehyde solution. A portion was stained with acridine orange, which caused bacteria to fluoresce under UV light so that they could be counted on a membrane filter. The quantity of soil used was determined by making bacterial counts over a range from 0.25 to 2 g per 1 l of 2% formaldehyde solution. Three samples from each horizon/site for a total of 18 counts (18 l of 2% formaldehyde) were examined. The resultant counts were in the $10^9/g$ range, which was consistent with other published data for soil bacteria. However, there was a significant variation in

data. For example, at the control site, organic horizon, 3 counts gave the following: mean = 72.1 bacteria on the membrane, SD = 8.1; mean = 52.9, SD = 10.1; mean = 43.8, SD = 6.2. The number of counts performed per sample was 10. However, it was estimated that, for a significance level of 0.05, it would be necessary to count 35 independent samples per site/horizon. Without added personnel, it was not possible to do 210 samples per date.

In addition, the methods of Tsai and Olsen (1991) to extract bacterial DNA from soil were used to estimate bacterial biomass . Quantification of extracted DNA was difficult to develop. As noted by others, including Tsai and Olsen (1991), DNA extracted from soil is contaminated with interfering substances. Direct measurement of DNA using a fluorometer were erratic due to soil contaminants, i.e. difficulty in repeat measuremens. We tried separating bacterial DNA from other soil substances by agarose gel electrophoresis, and then quantification of the DNA was attempted by UV fluorescence of ethidium bromide stained gels, with an "AMBIS" image analyzer. The data were too variable with this method. Further development of this method would have involved more extensive purification of soil DNA while preserving quantitatively accurate recovery, an uncertain outcome. Tsai and Olsen (1991) demonstrated that fungal DNA was not present in a large enough quantity to be a problem, and in the present work, electrophoresis gels failed to reveal eukaryotic, genome-size DNA. If eukaryotic, fungal DNA was degraded to prokaryotic size, this could provide an additional error to this method. This line

of research was discontinued.

2.3 Discussion.

Soil density measurements (Table 2) indicated that the organic horizon was approximately 3.65 times less dense (a very rough determination). Therefore, differences in amoeba counts between the organic and mineral horizons may be due in part to the difference in density between the horizons.

The pore structure of soil, as measured by soil suction, is an important factor in moisture retention in soil (Baver *et al.*, 1972) and represent the size of water-filled pores needed to support various sizes of microorganisms in soil (e.g. Derbyshire, 1975). Section 1 gives a description of the method and results of soil suction determinations. Assuming amoebae can occupy pores whose pore necks are 3 μm in diameter (Table 3), amoebae would require a moisture content in the organic horizon of 49% water and 10% water in the mineral horizon (Table 3) for growth. *In vitro* experiments indicate that *Acanthamoeba* can migrate through filters with 1.2 μm pores (Millipore Corp.) so that penetration through soil pores of 3 μm is not unreasonable. Soil moisture measurements of field samples were of the same order of magnitude as the soil suction determinations. However, specific moisture data, even taken from peak population samples, varied above and below the soil suction values of water content (Table 7).

Annual fluctuations of amoeba populations (vegetative and encysted organisms) (Figs. 5 to 8 and 10) represented a significant cycling of the population each season. Changes in

TABLE 7. Soil moisture (mean % w/w, \pm SD, n = 8) of organic and mineral horizons at the time of observed maximum annual population densities of amoebae.

Date	Organic horizon			Mineral horizon		
	CON	ANT	GND	CON	ANT	ORG
8/84	49 \pm 8	44 \pm	37 \pm	11 \pm 4	7 \pm 4	2 \pm 1
8/85	38 \pm 7	40 \pm	37 \pm	10 \pm 2	10 \pm	10 \pm 2
7/86	17 \pm 3	34 \pm	33 \pm	9 \pm 3	9 \pm 3	10 \pm 2
8/87	31 \pm 8	34 \pm	33 \pm	15 \pm 2	11 \pm 4	15 \pm 2
7/88	25 \pm 9	22 \pm	22 \pm	12 \pm 2	12 \pm 4	9 \pm 2
8/89	32 \pm 5	31 \pm	28 \pm	17 \pm 2	13 \pm 2	15 \pm 2
10/90	53 \pm 11	43 \pm	34 \pm	17 \pm 5	19 \pm 2	18 \pm 2
9/91	41 \pm 10	45 \pm	32 \pm	14 \pm 2	15 \pm 3	17 \pm 3
7/92	37 \pm 7	36 \pm	37 \pm	(not done)		

amoeba numbers may be due to the destruction of vegetative and encysted stages rather than food shortage since cysts are formed in response to starvation (Band, 1963). Cysts also decreased in number when the population collapsed in the autumn (Appendix Table C-2).

In addition to soil moisture, bacteria and fungi may regulate amoeba populations through their role as food (Singh, 1964) and toxin sources. As noted in the results (Section 2.2) fungal and actinomycete isolates served as food for amoebae as well as toxic, limiting factors in their survival. Clarholm (1981) studied changes in bacterial and protozoan populations in soil after a rain and concluded that amoebae were the primary regulators of bacterial populations in soil. *Acanthamoeba* can also use yeast as a food source (Heal, 1971). Derbyshire and Greaves (1967) found an increase in the number of amoebae in the rhizosphere as opposed to the surrounding soil but did not observe a qualitative difference in the type of amoebae. On the other hand, Geltzer (1963) observed a greater diversity of amoebae in the rhizosphere. Thus there is a significant interaction between amoebae and other soil microbes. Singh (1942) observed that *Rhizobium* was not eaten by amoebae; however, Danso and Alexander (1975) reported several genera of amoebae capable of feeding on *Rhizobium*. Singh (1941) found that the exo-toxin in *Serratia marcescens* prevented phagocytosis of bacteria by the amoebae. Singh (1945) found that chromogenic bacteria were toxic to amoebae. Thus some fungi and bacteria serve as food for amoebae while others are toxic.

At the time of annual population maxima there were no statistically significant differences between EM treatment and control sites. Few differences were noted at other times. Table 8 contrasts population maxima between pre- and post-operational years, and indicates no significant differences between these, except between the ground site and the control site mineral horizon, in which a small difference was observed. Table 9 contrasts cyst count maxima between pre- and post-operational years, and indicates no significant differences between these. Therefore, ELF electromagnetic radiation did not appear to have an effect on the growth of soil amoebae.

3. SPECIES CHARACTERIZATION

Species of soil amoebae present at the study sites were isolated and identified (Page, 1988) from soil enrichment plates described in Section 2. Based on morphology of vegetative amoebae and dormant cysts, no species differences were noted between sites. Species included: *Acanamoeba castellanii* Douglas, *A. polyphaga* Pushkarew, *A. astronyxis* Ray & Hayes, *Hartmannella* sp. Schaeffer, *Rosculus* sp. Hawes, *Naegleria gruberi* Schardinger, *Vahlkampfia* sp. Chatton & Lalung-Bonnaire, and *Mayorella* sp. Schaeffer. Rarely were exotic amoebae (e.g., *Leptomyxa reticulata* Goodey) were seen. Small, undescribed species were also observed; these were scored along with identified species for population size estimates (Section 2).

Other protozoa encountered included flagellates and *Colpoda* sp. Muller. Although testate amoebae, normally found in

TABLE 8. Pre- and post-operational comparison of population maxima. Data taken from Appendix Table C-1. The top table gives Pre-averages of maximum log count over the five years (1984-1988); the Post-averages are over four years (1989-1992) for the ORG horizon and over three years (1989-1991) for the MIN horizon.

TABLE OF MEANS AND STANDARD ERRORS

		Mean	SE
Antenna/ORG	Pre	5.03	0.50
	Post	4.79	0.30
Ground/ORG	Pre	5.04	0.50
	Post	4.82	0.25
Control/ORG	Pre	4.93	0.52
	Post	4.83	0.29
Antenna/MIN	Pre	4.62	0.58
	Post	4.14	0.47
Ground/MIN	Pre	4.67	0.55
	Post	3.99	0.39
Control/MIN	Pre	4.58	0.57
	Post	4.15	0.39

The post-averages are less than the Pre-averages reflecting a general decrease in maximum observed counts over the time period of the study.

BACI analysis (Stewart-Oaten et al., 1986) of annual log maximum counts. The following table gives the results of the tests of six contrasts. Each contrast compares a difference after the antenna was turned on (post) with the corresponding difference before the antenna was turned on (pre).

Comparison	Mean Diff. Before	Mean Diff. After	Difference t-value	p-value
A-C ORG	0.096	-0.038	-0.134	-0.798
G-C ORG	0.106	-0.010	-0.116	-0.940
A-G ORG	-0.010	-0.028	-0.018	-0.098
A-C MIN	0.032	-0.010	-0.042	-0.509
G-C MIN	0.082	-0.163	-0.245	-2.885
A-G MIN	-0.050	0.153	0.203	2.463

There was a marginally significant difference between and Ground and Control sites in the mineral horizon, the Ground site was slightly lower.

TABLE 9. Pre- and post-operational comparison of cyst count maxima. Data taken from Appendix Table C-2. The top table gives Pre-averages of maximum log count over the five years (1984-1988); the Post-averages are over four years (1989-1992) for the ORG horizon and over three years (1989-1991) for the MIN horizon.

TABLE OF MEANS AND STANDARD ERRORS

		Mean	SE
Antenna/ORG	Pre	4.02	0.33
	Post	4.49	0.14
Ground/ORG	Pre	4.04	0.36
	Post	4.56	0.29
Control/ORG	Pre	4.07	0.36
	Post	4.55	0.39
Antenna/MIN	Pre	3.51	0.33
	Post	3.96	0.36
Ground/MIN	Pre	3.56	0.36
	Post	3.80	0.26
Control/MIN	Pre	3.60	0.41
	Post	3.79	0.43

The post-averages are more than the Pre-averages reflecting a general increase in maximum observed counts over the time period of the study.

Before-After-Control-Impact (BACI) analysis (Stewart-Oaten et al., 1986) of annual log maximum cyst counts. The following table gives the results of the tests of six contrasts. Each contrast compares a difference after the antenna was turned on (post) with the corresponding difference before the antenna was turned on (pre).

Comparison	Mean Diff.	Mean Diff.	Difference	t-value	p-value
	Before	After			
A-C ORG	-0.046	-0.068	-0.022	-0.079	0.94
G-C ORG	-0.022	0.005	0.027	0.170	0.87
A-G ORG	-0.024	-0.073	-0.049	-0.234	0.82
A-C MIN	-0.094	0.170	0.264	1.342	0.23
G-C MIN	-0.044	0.013	0.057	0.397	0.71
A-G MIN	-0.050	0.157	0.207	1.102	0.31

No significant differences were observed Before (Pre) and After (Post) treatment.

freshwater, have been reported in soil (Darbyshire, 1975), these were not seen in the present study. Cellular slime molds, found in soil, were not observed, possibly because of the enrichment method. *Dictyostelium* Brefeld and other cellular slime molds need a solid substrate to support aggregation and spore formation; without this the amoeba stage cannot form spores for survival (Spudich, 1987). The enrichment method, described in Section 2, was based on a fluid phase over the substrate and would preclude spore formation.

Too few individuals of any amoeba species were present on each enrichment plate (Section 2) to quantify or to estimate species diversity without increasing the number of soil dilution plates significantly.

4. IN SITU GROWTH

Slime mold data obtained from the Wisconsin transmitter indicated a possible effect of the ELF radiation on the cell cycle (time between nuclear mitoses) of *Physarum* (Goodman, 1988). Therefore the *in situ* growth of amoebae in culture vessels buried in soil and exposed to the ELF antenna electromagnetic fields was monitored.

4.1 Methods.

The approach utilized clone isolates of *Acanthamoeba polyphaga* the study sites (Jacobson and Band, 1987). Direct counts of amoebae were made with a microscope to determine the increase in number of organisms. A log transform of these data provided a straight-line plot which were quantified by regression

analysis of growth rates (i.e., increase in number/unit time). Statistical comparisons between the growth rates were made with a t-test (Appendix Tables D-1, D-5, D-9). No differences were detected between the control, antenna and ground sites.

4.1.1 Rationale. Culture vessels were buried in soil at the sites and used electrodes to collect and distribute ELF induced soil currents through the culture saline. Direct measurement of amoeba growth was not possible (Section 2). Soil water is a saline suitable for amoeba growth, but it does not exist as a continuous aqueous phase in soil. Therefore soil exhibits a higher electrical resistance than would be the case for saline alone over a comparable distance, such as culture vessels, in which the saline is a continuous phase between the electrodes. In order to mimic the higher resistance found in soil in the continuous aqueous phase of a culture vessel containing saline, it would be necessary to dilute the saline below osmotic pressure and the ionic requirements required for growth of amoebae. Therefore, two different culture vessel configurations were used, one to mimic the voltage induced in soil by the ELF radiation (with a greater current, since the resistivity of saline is less than soil) and the other to mimic soil current (with a smaller voltage than observed in soil). Magnetic flux density is the same as the soil in all chambers. A summary of 76 Hz EM fields is given in Table 1.

4.1.2 Culture vessel. Personnel at the IIT Research Institute (IITRI) cooperated in the design and construction of electrical components used in the soil growth experiments. Their

procedures and diagrams are grouped at the end of Appendix D, Protocol, and include a test setup, determination of drive voltage, test cell hookup for matched E-field protocol, and test cell hookup for matched current density protocol.

Plastic t-tube connectors, used to connect water supplies, were used as culture vessels. They measured 11.5 cm long by 2 cm in diameter. The main axis of a t-tube was sealed at both ends with silicone stoppers that also held stainless steel, disc-shaped electrodes and insulated connectors fabricated by IITRI. The vertical arm was extended with a 15 cm long, 6mm diameter glass tube to provide a source of aeration and a sampling port. The glass tube was plugged with glass wool and extended up from the buried culture vessel to above soil level so that the gas phase was in equilibrium with air and to facilitate sampling. All components were sterilized before use. The culture vessels were filled to half their diameter, on the horizontal axis, with a sterile two-phase culture medium of 12.5 ml LSS-agar overlaid with 3.5 ml LSS, in which the amoeba and bacterial food were placed.

Lyophilized *E. coli* (Sigma Chem. Co., EC-11303), sterilized by ^{60}Co irradiation (282,000 R) at a concentration of 1 mg/ml was used as the food organism. This supported both maximum amoeba growth rate and maximum yield. Clone isolates of *A. polyphaga* from the study sites were used for the growth experiments. This species was chosen because its cyst morphology is distinctive and it is common to all sites. To count amoebae during growth, the culture vessel was agitated to suspend amoebae in the LSS. A

sample of the fluid phase containing amoebae was taken every 24 hr and fixed in 2% glutaraldehyde. Glutaraldehyde-fixed samples were counted with a hemacytometer in the lab. Known volumes were used for calculation of amoeba number per ml saline.

4.1.3 Data analysis. Data were obtained at each site from three cultures matched to electric fields found in the surrounding soil and from three cultures matched to the current density of the surrounding soil. Growth rates and regression calculations were determined over the period that samples were in exponential growth. The duration of exponential ranged from two to four days in duration (Appendix Figs. D-4, D-5, D-6). Analyses are presented in Appendix Tables D-1, D-5, D-9.

4.1.4 Between experiments. Cultures were left in the soil between experiments and then subcultured for use. In some cases the buried cultures became contaminated with a small flagellate, at which time new inoculum was made from one of the other replicates that had been exposed to similar electromagnetic fields.

At the end of the season, isoenzyme analyses (Jacobson and Band, 1987) were done on these amoebae. Section V gives a detailed description of allozyme methods. In the present experiments allozyme patterns were used to identify the amoeba clone used in the growth experiments. No change in isoenzyme pattern was observed between the original clone culture and subcultures grown in soil incubated at the sites. This was done to ensure that *A. polyphaga* from the surrounding soil did not invade the culture vessels and grow as contaminants.

4.2 Results.

Growth experiments in buried soil culture vessels were carried out in 1989, 1990, and 1991. The data are given in Appendix D. There were no significant site differences indicating that ELF EM exposure had no effect on the growth of *A. polyphaga* (Appendix Tables D-1, D-5, D-9).

4.3 Discussion.

Since the annual increase in the total soil amoeba population (Section 2) represented the cumulative growth of all the amoeba species in the population, this is comparable to the *in situ* growth experiments. In both cases (Section 2 and 4) ELF electromagnetic radiation had no effect on growth of amoebae. The data here in Section 4 provided a direct count of cultured amoebae, which was not possible to do directly with those in the soil (Section 2). Since the culture vessels were buried, amoebae growing in them were exposed to the same temperatures and magnetic fields acting as those in the surrounding soil.

5. GENETIC DIVERSITY

The objective of this study element was to monitor possible changes in genetic diversity at the antenna or ground sites relative to the control when the transmitter was operating at a full operational capacity.

If the electromagnetic field produced by the antenna affected organisms, this might be seen as some form of stress. Stress can be detected as a change in allozyme distribution (genetic diversity) in the population (Nevo *et al.*, 1977; Nevo,

1988; Parsons, 1987, 1989, 1992, 1993).

5.1 **Methods.** Clone isolates of *A. polyphaga* from the study sites were used for this analysis since they are easily identified by their cyst morphology (Page, 1988).

There were three limitations to this work:

1. No internal mobility controls were used.
2. Esterase patterns may be artifacts.
3. Absence of genetic recombination studies.

5.1.1 **Mobility controls.** Richardson et al. (1986) suggested the use of a mobility control such as a single isolate used in all electrophoresis studies. In the present study a common allele in each run was used as a reference point, an approach similar to Feder et al (1988). Pernin et al. (1992) studied the allozyme patterns of *Naegleria lovaniensis* clones from a natural population of this amoeba without the use of internal controls.

5.1.2 **Esterase allozymes.** The allozyme patterns obtained from esterases used in the present study were complex. This was not unique to the amoebae; similar patterns have been reported in other organisms (e.g. Harris and Hopkinson, 1978). Without genetic recombination studies on *A. polyphaga*, it is not possible to prove that clusters of enzyme bands are genetic loci nor is it possible to prove that the bands themselves represent alleles at a particular locus. Therefore, artifacts cannot be ruled out.

5.1.3 **Genetic recombination.** In the absence of genetic recombination studies (i.e. classical genetic crosses), there is no direct evidence that any of the allozyme patterns reported here represent genetic loci or alleles. The enzymes within a

cluster exhibit similar molecular features so that the assumption is made that these are allozymes at a genetic locus. However, some or all of these may be patterns due to artifacts, ranging from partial enzyme degradation to similar enzymes produced by unrelated genetic loci.

Cytologically, chromosome number, estimated from the metaphase plate, does not rule out polyploidy (Band and Mohrlok, 1973), although if this is the case, it certainly is not as extreme as the amoeba *Aulacantha* (Grell, 1953). Amoebae are at least 2C (diploid) judging from their allozyme patterns. If they are polyploid, this could alter allozyme patterns. Byers *et al.*, 1990), in a review article, speculated that *Acanthamoeba* is polyploid based on DNA content. This would also be true if the degree of polyploidy differed between clones.

5.2 Methods.

Clones were isolated from the plate cultures used to enumerate amoeba numbers, see Section 2.

The allozyme methods were developed for genetic analyses (Jacobson and Band, 1987) with field samples (5 clones from each study site) taken in 1985. These were used in 1986, 1987, 1988, before the antenna was in operation, and in 1991 and 1993 after the antenna was working. The allozyme analyses of genetic heterogeneity of *A. polyphaga*, were done in 1986, 1987, 1988, and 1991, using 10 clones from each site. For 1993, 30 clones were used from each site.

5.2.1 Collection and treatment

Collection methods are given in Section 2. Throughout each

growing season random samples were taken from each study site and amoebae were enumerated by a soil dilution method. From the enrichment plates, *A. polyphaga* was isolated, cloned, and used for the genetic diversity study. Although not all enrichment plates contained *A. polyphaga*, enough were present at the sampling dates to spread the samples over the collecting season for each year studied. For example, in 1993, 122 clones were isolated over the season, containing representative isolates from each study site. These were assigned number codes for identification, a blind experiment. During the course of allozyme analyses some clones were lost during culture while others were discarded when it was obvious that neither the allozyme patterns nor cyst morphology were of this species. From the original clone isolates, it was possible to obtain 30 clones from each site.

Starting in 1989, lyophilized *E. coli* (Sigma Chem. Co., EC-11303), sterilized by ^{60}Co irradiation (282,000 R), was used as food for growth of *A. polyphaga*, in place of living *E. coli* (K12) that was used before 1989. Therefore, the 1991 and 1993 enrichments were done with lyophilized *E. coli*.

Amoeba clone isolates (approximately $2 \times 10^7/\text{ml}$), separated from bacteria by centrifugation, were frozen over liquid N₂ in 200 μl of lysis buffer (10 mM Tris, 1 mM EDTA and 0.5 mM NADP, pH 6.8) (Selander *et al.*, 1986). The lysate was thawed, diluted with 200 μl of a mixture of glycerol and electrophoresis running buffer (pH 8.2) and centrifuged to remove debris. Electrophoresis was done with 50 μl aliquots.

Polyacrylamide gel electrophoresis was used (Jacobson and Band, 1987) with the running gel at pH 8.2. Acrylamide reagents were obtained from Boehringer Mannheim Corp., while buffers and staining reagents were obtained from Sigma Chemical Co. Data analysis procedures were those given in (Jacobson & Band, 1987).

5.2.2 Allozyme Methods. The preliminary study done in 1985 (Jacobson and Band, 1987) utilized propionyl esterase, acetyl esterase, and tetrazolium oxidase (= superoxide dismutase), all of which are shown in Table 10; the allozymes used and the number of loci analyzed (with references) are also given. The specific allozymes used differed between the 1986-1988 group and the 1991 and 1993 groups. Since the data from the sites were the same (Table 10), differences in the choice of allozymes did not affect results between years.

The allozyme banding patterns in polyacrylamide gels differed in intensity between loci for many allozymes. It has been noted (Selander et al., 1986) that allozyme activity, reflected by differences in staining intensity, is influenced by growth conditions, although not in an alteration of banding patterns. All amoebae were grown under the same conditions to avoid stain intensity differences.

In 1985 we isolated 5 clones of *A. polyphaga* per site, without regard to horizon, froze them in saline (Daggett and Nerad, 1983), and performed allozyme tests using three enzymes and a total of 10 loci (Jacobson and Band, 1987). Although many enzymes worked with axenic amoeba cultures (e.g. Daggett and Nerad, 1983; Pernin et al., 1985), this was not the case for

TABLE 10. Allozymes used for loci

Enzymes	year					Stain References*
	1986	1987	1988	1991	1993	
lactate dehydrogenase (LDH)	+	+	+	+	+	4
L-threonine dehydrogenase (LTD)	+	+	+	+	+	3
isocitrate dehydrogenase (ICD)	+	+	+	-	-	3
hexokinase (HK)	+	+	+	-	-	4
propionyl esterase (pH 5.7)(PE)	+	+	+	+	+	4
butyryl esterase (pH 5.7)(BE)	+	+	+	+	+	1
acetyl esterase (AE)	+	+	+	+	+	4
phosphoglucomutase (PGM)	+	+	+	+	+	4
6-phosphogluconate dehydrogenase (PGM)	+	-	-	-	-	1
Bhydroxybutyrate dehydrogenase (BDH)	+	-	-	-	-	1
malic enzyme (ME)	+	-	-	-	-	4
leucine aminopeptidase (LAP)	+	-	-	-	-	4
glutamate dehydrogenase (GDH)	+	-	-	+	+	4
arginine amino peptidase (AAP)	+	-	-	-	-	1
superoxide dismutase (SOD)	+	-	-	+	+	2
acid phosphatase (AP)	-	-	-	+	+	4
number of loci	30-34	27	28			

- * 1. Daggett, P. & Nerad, T.A. (1983). Procedures for isoenzyme electrophoretic analysis. American Type Culture Collection, 2nd ed.
2. Murphy, R.S., Sites, Jr., J.W., Buth, D.G. & Christopher, H.H. (1990). Proteins I: Isozyme Electrophoresis, Ch. 4., Molecular Systematics (Hillis, D.M. & Moritz, C., eds.) Sinauer Associates, Inc.
3. Pernin, P., Cariou, M.-L. & Jacquier, A. (1985). Biochemical identification and phylogenetic relationships in free-living amoebas of the genus Naegleria. J. Protozool. 32, 592-603.
4. Werth, C.R. (1985). Implementing an isozyme laboratory at a field station. Virginia J. Sci. 36, 53-76.

amoebae grown on bacteria. Starting in 1986 the allozyme method was improved by freezing cells in a stabilizing medium (Selander et al., 1986), which permitted running a variety of allozymes. The 1986/1987 annual report cited a drop in genetic diversity between 1985 and 1986, which was attributed to *A. polyphaga*'s response to drought. However, the genetic diversity values obtained with larger sample sizes and more loci indicated similar values across subsequent years (1986, 1987, 1988, 1991 and 1993). Therefore, the drop in diversity between 1985 and 1986 may have been due to larger sample sizes and better techniques used after 1985.

5.3 Results

Allozyme data among research sites (Table 11) did not differ between EM exposure regimes for the years studied i.e., 1986 to 1988, 1991 and 1993 (Table 12). In the analysis given in Table 11, for 1986, 1987, 1988 and 1991, with ten (10) isolates, there were 45 pairs of isolates. Nei's D genetic distance was calculated for each pair, and the mean D was calculated for the antenna, ground and control sites. The differences in the means were tested for statistical significance. For the BACI analyses (Stewart-Oaten et al., 1986) of "average Nei distance", the Before period consisted of the years 1986-1988 and the After period consisted of the years 1991 and 1993 with the two measurements for 1991 at the Impact (antenna) site averaged for input into the analysis.

The independence of the 45 pairs of isolates from each other was also examined. On analysis, for each site, the 45 distances

TABLE 11. Genetic diversity expressed as genetic distance (means \pm SD) in each site, 1986 to 1993, with 1985 data added from Jacobson & Band (1987).

1. Summary data:

year	1986	1987	1988	1991	1993
clones/site	10	10	10	10	30

number of Nei's genetic distance determinations/site (n):

	45	45	45	45	435
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SITE	Mean genetic distance \pm Std. Dev.				
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Control	0.5108 \pm 0.175	0.5494 \pm 0.175	0.5643 \pm 0.210	0.5713 \pm 0.228	0.5549 \pm 0.1794
Antenna	0.5589 \pm 0.178	0.5558 \pm 0.198	0.5333 \pm 0.147	0.4459 \pm 0.1562	0.5201 \pm 0.1448
				0.3154* \pm 0.119	
Ground	0.5314 \pm 0.186	0.5229 \pm 0.167	0.5026 0.129	0.5878 \pm 0.168	0.5538 \pm 0.1586

2. Preliminary data obtained in 1985 (Jacobson & Band, 1987) with 5 clones/site, and 10 genetic comparisons/site (n = 10):

Control: 1.1452 \pm 0.565

Antenna: 1.0576 \pm 0.521

Ground: 1.1452 \pm 0.444

*A second set of data from the antenna site in 1991.

TABLE 12. Pre- and Post-operational comparisons of average Nei distance. Data taken from Table 11. The top table gives the Pre-averages of average Nei distance over the three years (1986-1988) and the Post-averages over the two years 1991 and 1993. The two measurements for 1991 at the Antenna site were averaged.

TABLE OF MEANS AND STANDARD ERRORS

	CONTROL		ANTENNA		GROUND	
Year	Mean	SE	Mean	SE	Mean	SE
1986	0.511	0.026	0.559	0.025	0.531	0.027
1987	0.549	0.051	0.556	0.026	0.523	0.054
1988	0.564	0.090	0.533	0.040	0.503	0.036
1991	0.571	0.102	0.381	0.036	0.588	0.047
1993	0.555	0.035	0.520	0.027	0.554	0.027

		Mean	SE
Antenna	Pre	0.549	0.054
	Post	0.451	0.045
Ground	Pre	0.519	0.070
	Post	0.571	0.054
Control	Pre	0.541	0.107
	Post	0.563	0.108

BACI ANALYSIS^a OF AVERAGE NEI DISTANCE

Comparison	Mean	Diff.	Mean	Diff.	Difference	t-value	p-value
ANT-CON	0.008		-0.113		0.121	1.858	0.16
GND-CON	-0.022		0.008		-0.030	-0.978	0.40
ANT-GND	0.030		-0.121		-0.151	2.338	0.10

No significant differences were observed for Pre- (Before) and Post- (After) the antenna was turned on.

^aStewart-Oaten et al. (1986).

could not be regarded as a sample of 45 independent measurements because of the correlation for the differences corresponding to pairs with a common isolate. To estimate the correlation of measurements that share data from a common isolate, we examined all 990 differences of the 45 distances, taken two at a time. There are 630 pairs that did not share a common isolate and 360 pairs did. Assuming a common correlation coefficient p for pairs which share a common isolate, the sample variance of the 630 differences is an estimate of $2 \sigma^2$, and the sample variance of the 360 differences is an estimate of $2(1 - p)\sigma^2$. An estimate of the correlation coefficient p is derived from the ratio of the sample variances. With the above correlation structure in the summands in the average of 45 distances, the standard error formula is $[1 + 16p]^{1/2} \sigma/\sqrt{45}$ and not the familiar $\sigma/\sqrt{45}$.

For 1993, with 30 isolates, there were 435 pairs of isolates. Nei's D genetic distance was calculated for each pair, and the mean D was calculated for the antenna, ground and control sites. The differences in the means were tested for statistical significance. For each site, the 435 distances cannot be regarded as a sample of 435 independent measurements because of the correlation of measurements that share data from a common isolate. To estimate this correlation, we examined all 94,395 differences of the 435 distances, taken two at a time. There are 82,215 pairs that do not share a common isolate and 12,180 pairs that do. Assuming a common correlation coefficient p for pairs sharing a common isolate, the sample variance of the 82,215 differences is an estimate of $2 \sigma^2$ and the sample variance of the

12,180 differences is an estimate of $2(1 - p)\sigma^2$. An estimate of the correlation coefficient p is derived from the ratio of the sample variances. With the above correlation structure in the summands in the average of 435 distances, the standard error formula is $[1 + 56p]^{1/2} \sigma/\sqrt{435}$ and not the familiar $\sigma/\sqrt{435}$.

For the years 1988, 1991, and 1993 the estimated correlation coefficients ranged from 0.146 to 0.407 with an average of 0.241, confirming that the standard error formula σ/\sqrt{n} will underestimate the true standard error. However, the 1986 and 1987 data showed less correlation.

Table 12 gives mean Nei D and the standard error of the mean for each year and site. Pre- and post-means are averages over the years 1986, 1987 and 1988 and over the years 1991 and 1993, respectively. The standard errors of all contrasts were calculated by the square root of added variances. The z-statistic is the contrast divided by its standard error.

Site to site comparisons , using the BACI analysis failed to reveal differences before and after the antenna became operational.

As a supplement to Tables 11 and 12, Appendix Table E-1 to E-3 gives the allele values used to calculate Nei's values in 1993, while a summary of Nei's genetic distance values for 1993 is given in Appendix Table E-4 to E-6.

5.4 Discussion.

There is no evidence for sexual reproduction *in vitro* among amoebae commonly isolated, but this does not preclude the possibility of genetic recombination in the soil habitat. In

another genus, *Naegleria*, allozyme patterns in several species has been used to speculate on the presence of genetic recombination (Cariou and Pernin, 1987). An allozyme study of *N. lovaniensis* isolates from natural habitats (Pernin, et al., 1992) is consistent with my data. Laboratory stability of *N. lovaniensis* supported asexual reproduction and no genetic recombination in clone isolates maintained in the laboratory (Pernin et al., 1992). Allozyme patterns of clones isolated from the wild were rarely shared in *Naegleria*, for the most part each clone isolate was different from the others. This was also the case for *A. polyphaga*. The stability of allozyme patterns over time for clonal isolates of *A. polyphaga* (Table 13) demonstrated the absence of genetic recombination *in vitro*. Given the significant annual cycling of population size each year (see Section 2), this fluctuation might provide a selection mechanism *for affecting genetic diversity. However the population size of amoebae at the start of each growing season, approximately $10^3/g$ soil, was too large to consider it to be a genetic bottleneck.

In conclusion, the genetic diversity data from site to site and from year to year were similar. The data obtained were similar to that obtained by Pernin et al. (1992) with the amoeba *N. lovaniensis*. Again genetic crosses have not been possible with *N. lovaniensis*, although Pernin's data have been used to propose the existence of sexual reproduction in the natural habitat (Cariou and Pernin, 1987; Pernin et al., 1992). Therefore, the absence of classical genetic inheritance patterns for *A. polyphaga* limits the usefulness of allozyme data in attempting to interpret patterns as alleles of genetic loci.

TABLE 13. Allozyme stability, samples of clones isolated 21 July 93 (63, 64) and 7 Sep 93 (120) were frozen for allozyme analysis. Continuous cultures from these clones (63s, 64s & 120s) were maintained to 1 Mar 94 and then frozen for allozyme analysis. Clones 63 and 64 were isolated from the ground site, organic horizon, from separate culture enrichment plates; clone 120 was isolated from the antenna site, mineral horizon.

Alleles for Given Clones			
Loci	63/63s	64/64s	120/120s
AE1	1/2, 1/2	1, 1	1/2, 1/2
2	1, 1	1, 1	1/2, 1/2
3	1/2, 1/2	1/2, 1/2	1/5, 1/5
4	1/3, 1/3	1/2, 1/2	1/2, 1/2
PE1	1, 1	1, 1	3/4, 3/4
2	1, 1	1/2, 1/2	1/2, 1/2
3	4, 4	3, 3	1/2, 1/2
4	1/2, 1/2	1/3, 1/3	1/3, 1/3
5	1/2, 1/2	3/4, 3/4	1/2, 1/2
BE1	2, 2	1/2, 1/2	1/2, 1/2
2	1, 1	1, 1	1/2, 1/2
3	2, 2	1/2, 1/2	1/2, 1/2
4	1/2, 1/2	1, 1	1, 1
5	1/2, 1/2	1/2, 1/2	1/2, 1/2
SOD1	1/3, 1/3	1/3, 1/3	1, 1
2	1/3, 1/3	1, 1	3/4, 3/4
3	1/2, 1/2	1/4, 1/4	1/2, 1/2
ACP1	1/2, 1/2	1/2, 1/2	1/4, 1/4
2	1/3, 2	1/3, 1/3	4, 4
3	1/3, 1/3	1/2, 1/2	1/4, 1/4
GDH1	1, 1	1/5, 1/5	1/2, 1/2
LTD1	1/2, 1/2	1/3, 1/3	3/4, 3/4
2	1/2, 1/2	1/3, 1/3	1/2, 1/2
LDH1	1/2, 1/2	1/3, 1/3	1/3, 1/3
2	1/2, 1/2	3, 3	1/4, 1/4
PGM1	1, 1	1, 1	1, 1
2	1, 1	1, 1	1, 1
3	1/2, 1/2	2, 2	2, 2

LITERATURE CITED

- Band, R.N. (1963). Extrinsic requirements for encystation by the soil amoeba, *Hartmannella rhysodes*. *J. Protozool.* 10, 101-106.
- Band, R.N. and Mohrlok, S. (1969). The respiratory metabolism of *Acanthamoeba rhysodes* during encystation. *J. Protozool.* 59, 351-358.
- Band, R.N. and Mohrlok, S. (1973). The cell cycle and induced amitosis in *Acanthamoeba*. *J. Protozool.* 20, 654-657.
- Baver, L.D., Gardner, W.H. and Gardner, W.R. (1972). *Soil Physics* (4th ed.), Wiley, New York 498 pp.
- Bryant, R.J., Woods, L.E., Coleman, D.C., Fairbanks, B.C., McClellan, J.F. and Cole, C.V. (1982). Interactions of bacterial and amoeba populations in soil microcosms with fluctuating moisture content. *Appl. Environ. Microbiol.* 43, 747-752.
- Byers, T.J., Hugo, E.R. and Stewart, V.J. (1990). Genes of *Acanthamoeba*:DNA, RNA and Protein Sequences (A Review). *J. Protozool.* 37, 17s-25s.
- Cariou, M.L. and Pernin, P. (1987). First evidence for diploidy and genetic recombination in free-living amoebae of the genus *Naegleria* on the basis of electrophoretic variation. *Genetics* 115, 265-270.
- Clarholm, M. (1981) Protozoan grazing of bacteria in soil--impact and importance. *Microb Ecol* 7, 343-350.

- Daggett, P. and Nerad, T.A. (1983). Procedures for isoenzyme electrophoretic analysis. American Type Culture Collection, 2nd ed.
- Danso, S.K.A. and Alexander, M. (1975). Regulation of predation by prey density: the protozoan-Rhizobium relationships. Appl. Microbiol. 29, 515-21.
- Darbyshire, J.F. (1975). Soil Protozoa--Animalcules of the Subterranean Microenvironment. In: *Soil Microbiology*, N. Walker (ed.) Ch. 8, Wiley & Sons, New York.
- Darbyshire, J.F. and Greaves, M.P. (1967). Protozoa in the rhizosphere of *Sinapis alba L.*, *Trifolium reptans L.*, and *Lolium perenne L.* Canad. J. Microbiol. 13, 1075-68.
- Darbyshire, J.F., Wheatley, R.E., Graves, M.P. and Inkson, R.H.E. (1974). A rapid micromethod for estimating bacterial and protozoan populations in soil. Rev. Ecol. Sol 11, 465-475.
- Elliott, E.T., Anderson, R.V., Coleman, D.C. and Cole, C.V. (1980). Habitable pore space and microbial trophic interactions. Oikos 35, 327-335.
- Feder, J.L., Chilcote, C.A. and Bush, G.L. (1988). Genetic differentiation between sympatric host races of the apple maggot fly *Rhagoletis pomonella*. Nature 336, 61-64.
- Fisher, R.A. and Yates, F. (1963). *Statistical Tables in Biological, Agricultural and Medical Research*, Oliver and Boyd, Edinburgh.
- Geltzer, J.D. (1963). On the behavior of soil amoebae in the rhizosphere of plants. Pedobiologia 2, 249-251.

- Goodman, E.M. (1988). The effects of exposing the slime mold *Physarum polycephalum* to electromagnetic fields. In: *Compilation of 1987 Annual Reports of the Navy Communication System Ecological Monitoring Program*. IIT Research Institute, Chicago.
- Grell, K.G. (1953). Die chromosomen von *Aulacantha scolymantha* Haeckel. Arch. Protistenk. 99, 1-54.
- Hanssen, J.F., Thingstad, T.F. and Goksoyr, J. (1974). Evaluation of hyphal lengths and fungal biomass in soil by a membrane filter technique. Oikos 25, 102-107.
- Haradem, D.P., Gauger, J.R. and Zapotosky, J.E. (1994). ELF Communications System Ecological Monitoring Program: Electromagnetic Field Measurements and Engineering Support-- Final Report. IIT Research Institute, Technical Report D06209-1, Contract No. N00039-93-C-001.
- Harris, H. and Hopkinson, D.A. (1978). *Handbook of Enzyme Electrophoresis in Human Genetics*. North Holland Publ. Co., Amsterdam.
- Heal, O.W. (1970). Methods of study of soil protozoa. In: *Methods of Study in Soil Ecology*, J. Phillipson (ed.). UNESCO, 119-126.
- Heal, O.W. (1971). Protozoa. In: *Methods of Study in Quantitative Soil Ecology: Population, Production and Energy Flow*. J. Phillipson (ed.), Blackwell Scientific Publ., Oxford, pp. 51-71.

- Hobbie, J.E., Daley, R.J. and Jasper, S. (1977). Use of nucleopore filters for counting bacteria by fluorescence microscopy. *Appl. and Environ. Microbiol.* 33, 1225-1228.
- Jacobson, L. and Band, R. N. (1987). Genetic heterogeneity in a natural population of *Acanthamoeba polyphaga* from soil, an isoenzyme analysis. *J. Protozool.* 34, 83-86.
- Nevo, E., Shimony, T. and Lbini, M. (1977). Thermal selection of allozyme polymorphisms in barnacles. *Nature* 267, 699-701.
- Nevo, N. (1988). Genetic diversity in nature, patterns and theory. *Evol. Biol.* 23, 217-246.
- Page, F.C. (1988). *Freshwater and Soil Gymnamoebae*. Freshwater Biological Assoc., The Ferry House, Ambleside, Cumbria, U.K.
- Parsons, P.A. (1987). Evolutionary rates under environmental stress. *Evol. Biol.* 21, 311-347.
- Parsons, P.A. (1989). Environmental stress and conservation of natural populations. *Ann. Rev. Ecol. Syst.* 20, 29-49.
- Parsons, P.A. (1992). Evolutionary adaptation and stress, the fitness gradient. *Evol. Biol.* 26, 191-223.
- Parsons, P.A. (1993). Evolutionary responses to environmental stress, a symposium. *Amer. Naturalist* 142 Supplement.
- Pernin, P., Atarya, A. & Cariou, M.L. (1992). Genetic structure of natural populations of the free-living amoeba, *Naegleria lovaniensis*. Evidence for sexual reproduction. *Heredity* 68, 173-181.
- Pernin, P., Cariou, M.L. & Jacquier, A. (1985). Biochemical identification and phylogenetic relationships in free-living amoebas of the genus *Naegleria*. *J. Protozool.* 32, 592-603.

- Richards, L.A. (1954). *Diagnosis and Improvement of Saline and Alkali Soils.* Agriculture Handbook No.60, USDA.
- Richardson, B.J., Baverstock, P.R. and Adams, M. (1986). *Allozyme electrophoresis: A handbook for animal systematics and population studies,* Academic Press, Inc., Sydney.
- Selander, R.K., Caugant, D.A., Ochman, H., Musser, J.M., Gilmour, M.N. and Whittam, T.S. (1986). Methods of multilocus enzyme electrophoresis for bacterial population genetics and systematics. *Appl. and Environ. Microbiol.* 51, 873-884.
- Singh, B.N. (1941). Selectivity in bacterial food by soil amoebae in pure mixed culture and in sterilized soil. *Ann. Appl. Biol.* 28, 52-64.
- Singh, B.N. (1942). Selection of bacterial food by soil flagellates and amoebae. *Ann. Appl. Biol.* 29, 18-22.
- Singh, B.N. (1945). The selection of bacterial food by soil amoebae, and the toxic effects of bacterial pigments and other products on soil protozoa. *Brit. J. Exp. Pathol.* 26, 316.
- Singh, B.N. (1946). A method of estimating the numbers of soil protozoa, especially amoebae, based on their differential feeding on bacteria. *Ann. Appl. Biol.* 33, 112-119.
- Singh, B.N. (1964). Soil protozoa and their probable role in soil fertility. *Bull. Natl. Inst. Sci., India*, No. 26, 238-244.
- Spudich, J.A. 1987. *Methods in Cell Biology*, vol. 28 Academic Press, Orlando, Florida.

Stewart-Oaten, A., Murdoch, W.W. and Parker, K.R. 1986.

Environmental impact assessment: "pseudoreplication" in time. Ecology 67, 929-940.

Tsai, Y-L and Olson, B.H. (1991). Rapid method for direct extraction of DNA from soil and sediments. Appl. Environ. Microbiol. 57, 1070-1074.

Wright, R. T. and Coffin, R. B. (1984). Measuring microzooplankton grazing on planktonic marine bacteria by its impact on bacterial production. Microb. Ecol. 84, 137-149.

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APPENDIX A
Maps of Study Sites

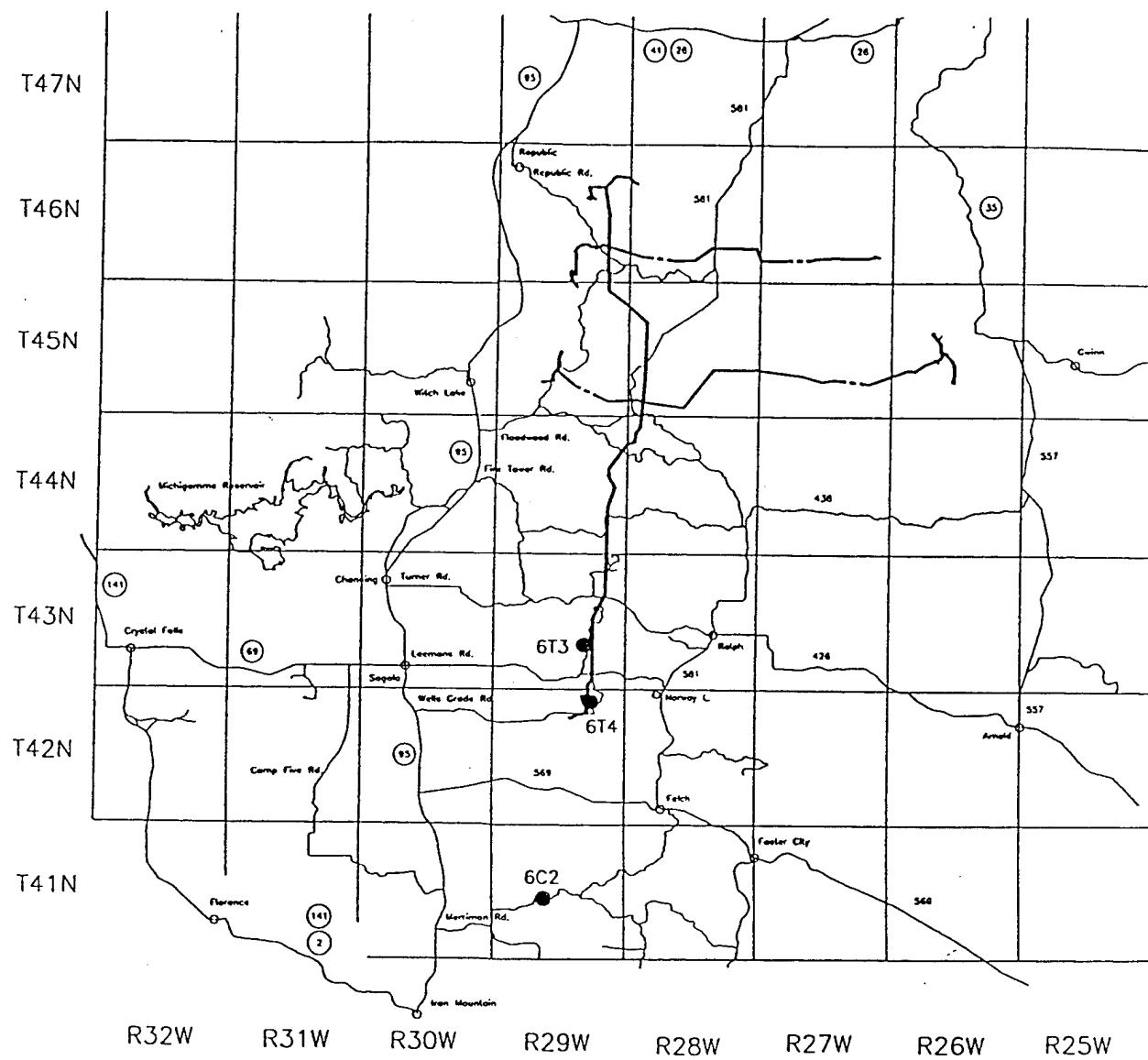


FIGURE A-1. Site map (Haradem et al., 1994): Control site (6C2), Merriman Truck Trail; Antenna site (6T3), Leeman's Rd., Ground site (6T4), Wells Grade Rd.

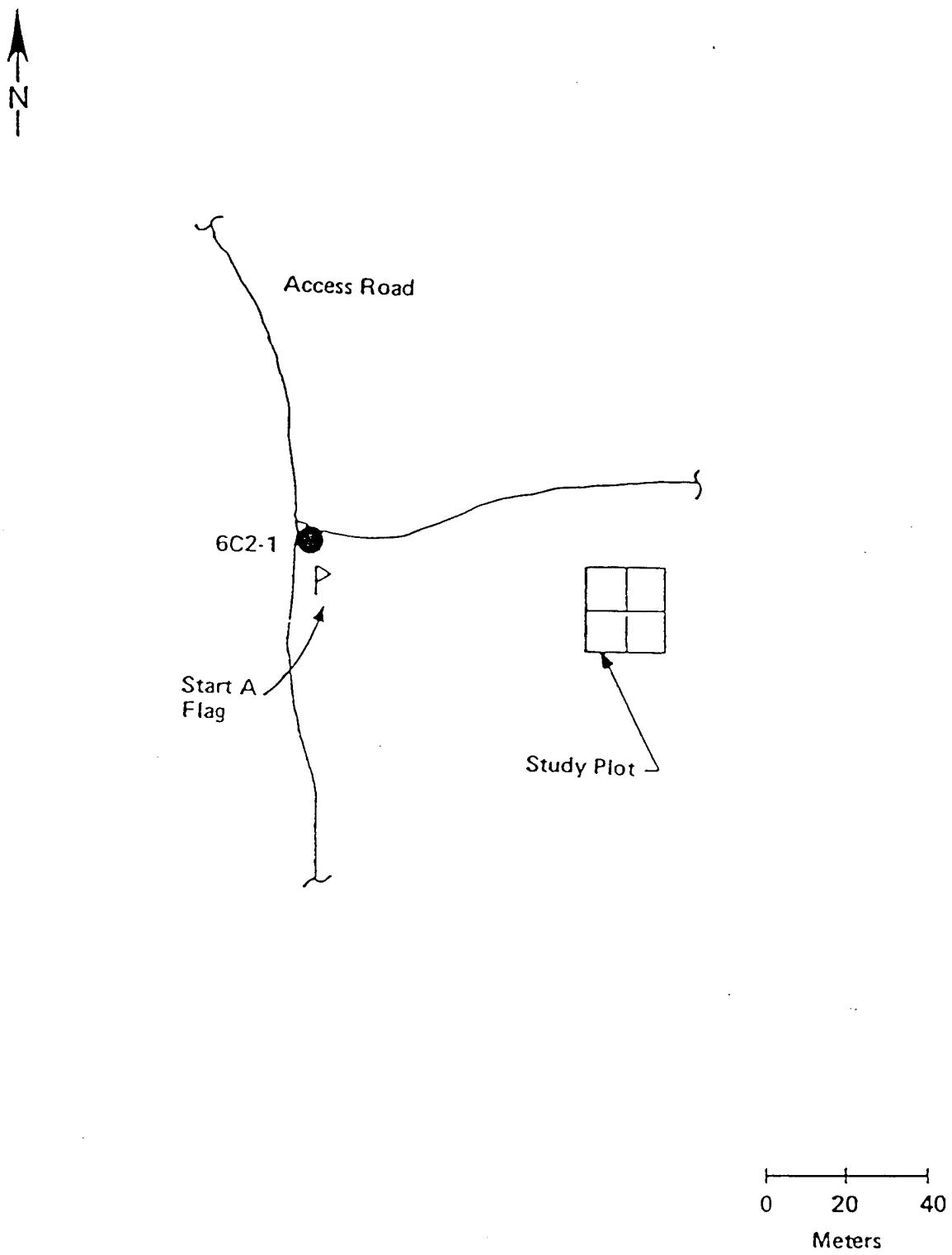


FIGURE A-2. Control site (6C2), Merriman Truck Trail (Haradem et al., 1994).

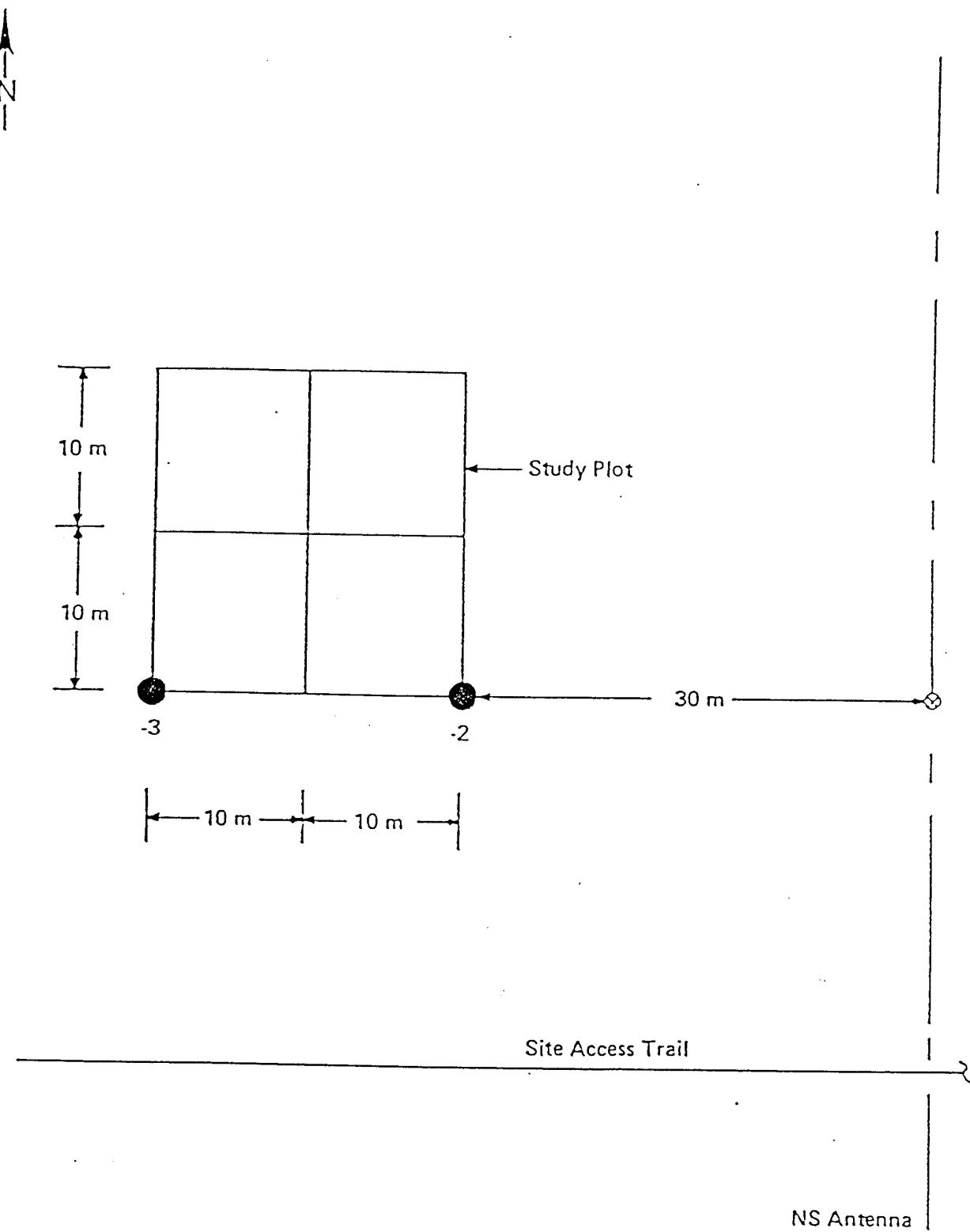


FIGURE A3. Antenna site (6T3), Leeman's Rd. (Haradem et al., 1994).

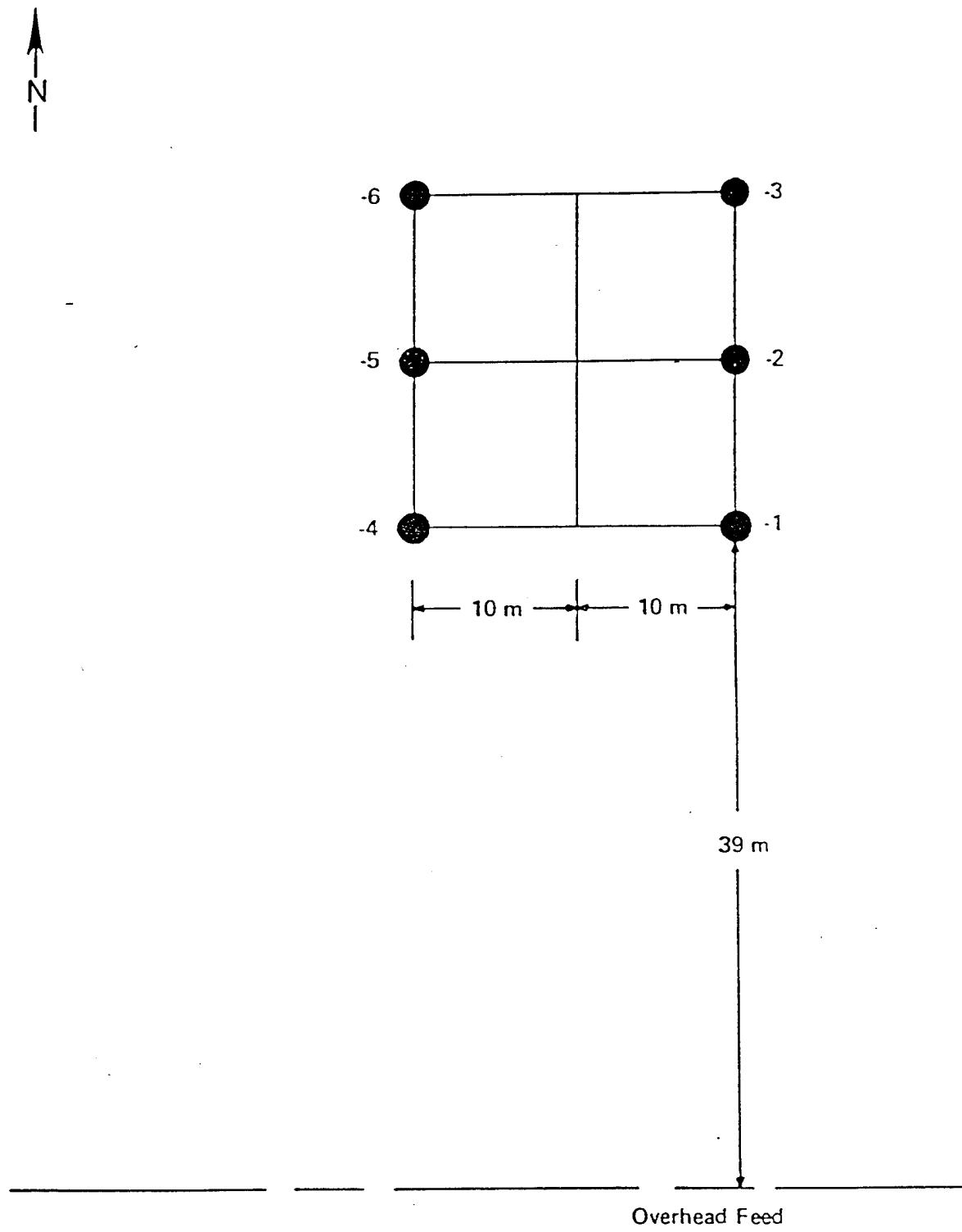


FIGURE A-4. Ground site (6T4), Wells Grade Rd. (Haradem et al., 1994).

APPENDIX B
Soil Moisture

TABLE B-1. SOIL MOISTURE (mean %, w/w \pm SD, n = 8) 1984.

HORIZON	CONTROL SITE		ANTENNA SITE		GROUND SITE	
	ORGANIC	MINERAL	ORGANIC	MINERAL	ORGANIC	MINERAL
DATE						
6/18	52 \pm 6	16 \pm 2	46 \pm 7	9 \pm 3	50 \pm 10	17 \pm 5
6/27	53 \pm 7	16 \pm 2	46 \pm 11	12 \pm 3	52 \pm 9	15 \pm 2
7/9	37 \pm 16	16 \pm 2	36 \pm 12	8 \pm 2	34 \pm 4	11 \pm 2
7/18	78 \pm 8	42 \pm 3	79 \pm 13	35 \pm 2	79 \pm 7	32 \pm 3
8/6	49 \pm 8	11 \pm 4	44 \pm 10	7 \pm 4	37 \pm 13	12 \pm 1
9/3	48 \pm 12	17 \pm 1	33 \pm 10	12 \pm 3	35 \pm 6	12 \pm 2
10/2	48 \pm 5	18 \pm 3	32 \pm 5	13 \pm 1	43 \pm 9	16 \pm 3

ONE-WAY ANOVA (between sites):

Date	ORGANIC			MINERAL	
	D.F.	M.S.	F =	D.F.	M.S.
6/18	Between	2	152	2	152
	Within	21	12.7	21	12.7
	F =		1.21 (NS)		12**
6/27	Between	2	114.7	2	34.6
	Within	21	83.7	21	5.7
	F =		1.37 (NS)		6.12**
7/9	Between	2	18.7	2	130.7
	Within	21	138.7	21	4
	F =		0.13 (NS)		32.67**
7/18	Between	2	2.7	2	210.7
	Within	21	94	21	7.3
	F =		0.03 (NS)		28.73**
8/6	Between	2	290.7	2	28.7
	Within	21	111	21	56
	F =		2.62 (NS)		5.09*
9/3	Between	2	530.7	2	66.7
	Within	21	93.3	21	4.7
	F =		5.69*		14.29**
10/2	Between	2	536	2	50.7
	Within	21	43.7	21	6.3
	F =		12.27**		8**

* 5% significance level

** 1% significance level

TABLE B-1 cont'd (1984)

Bonferroni paired t-tests for dates with significant ANOVA differences, 1984:

DATE	HORIZON	CON/ANT	CON/GND	ANT/GND
6/10	MINERAL	**(p<.01)	NS(p>.05)	**(p<.01)
6/27	"	*(p<.05)	NS(p>.05)	NS(p>.05)
7/9	"	***(p<.001)	***(p<.001)	*(p<.05)
7/18	"	***(p<.001)	***(p<.001)	NS(p>.05)
8/6	"	NS(p>.05)	NS(p>.05)	*(p<.05)
9/3	ORGANIC	***(p<.001)	***(p<.001)	NS(p>.05)
9/3	MINERAL	**(p<.01)	**(p<.01)	NS(p>.05)
10/2	ORGANIC	***(p<.001)	NS(p>.05)	*(p<.05)
10/2	MINERAL	**(p<.01)	NS(p>.05)	NS(p>.05)

TABLE B-2. SOIL MOISTURE (mean %, w/w \pm SD, n = 8) 1985.

HORIZON	CONTROL SITE		ANTENNA SITE		GROUND SITE	
	ORGANIC	MINERAL	ORGANIC	MINERAL	ORGANIC	MINERAL
DATE						
6/10	41 \pm 7	17 \pm 1	42 \pm 8	11 \pm 1	46 \pm 9	12 \pm 2
6/28	44 \pm 6	16 \pm 3	48 \pm 14	13 \pm 3	46 \pm 15	14 \pm 2
7/22	30 \pm 3	8 \pm 0.7	26 \pm 6	7 \pm 2	26 \pm 8	7 \pm 2
8/20	38 \pm 7	10 \pm 2	40 \pm 4	10 \pm 2	38 \pm 7	10 \pm 2
9/6	35 \pm 7	13 \pm 1	32 \pm 6	14 \pm 3	31 \pm 5	13 \pm 1
10/5	61 \pm 7	21 \pm 2	45 \pm 13	14 \pm 3	57 \pm 10	16 \pm 3

ONE-WAY ANOVA:

DATE	ORGANIC			MINERAL	
	D.F.	M.S.	F =	D.F.	M.S.
6/10	Between	2	63.4	2	77.3
	Within	21	66.5	21	1.9
			0.95 (NS)		41.46**
6/28	Between	2	26.3	2	13.1
	Within	21	163.7	21	7.8
			0.16 (NS)		1.68 (NS)
7/22	Between	2	43.4	2	1.1
	Within	21	34.5	21	2.7
			1.26 (NS)		0.42 (NS)
8/20	Between	2	5.8	2	0.8
	Within	21	30.5	21	4.1
			0.19 (NS)		0.18 (NS)
9/6	Between	2	35.1	2	0.1
	Within	21	35.6	21	3.5
			0.99 (NS)		0.03 (NS)
10/5	Between	2	556.5	2	104.9
	Within	21	106.3	21	9.4
			5.24*		11.22**

* 5% significance level

** 1% significance level

TABLE B-2 cont'd (1985)

Bonferroni paired t-tests for dates with significant ANOVA differences, 1985:

DATE	HORIZON	CON/ANT	CON/GND	ANT/GND
6/10	MINERAL	***(p<.001)	***(p<.001)	NS(p>.05)
10/5	ORGANIC	*(p<.05)	NS(p>.05)	NS(p>.05)
10/5	MINERAL	***(p<.001)	**(p<.01)	NS(p>.05)

TABLE B-3. SOIL MOISTURE (mean %, w/w \pm SD, n = 8) 1986.

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HORIZON	CONTROL SITE		ANTENNA SITE		GROUND SITE	
	ORGANIC	MINERAL	ORGANIC	MINERAL	ORGANIC	MINERAL
DATE						
6/16	39 \pm 10	15 \pm 2	36 \pm 11	12 \pm 2	27 \pm 7	13 \pm 2
7/23	17 \pm 3	9 \pm 3	35 \pm 9	9 \pm 3	34 \pm 8	10 \pm 2
8/21	28 \pm 10	11 \pm 3	35 \pm 12	9 \pm 4	39 \pm 8	11 \pm 2
9/13	43 \pm 15	17 \pm 4	56 \pm 4	15 \pm 4	35 \pm 10	14 \pm 2
10/14	56 \pm 8	23 \pm 4	43 \pm 7	19 \pm 5	58 \pm 8	22 \pm 3

ONE-WAY ANOVA:

DATE	ORGANIC		MINERAL	
	D. F.	M. S.	D.F.	M. S.
6/16	Between	2	250.6	2
	Within	21	87.1	21
	F =		2.88*	3.11 (NS)
7/23	Between	2	763.6	2
	Within	21	52.2	21
	F =		7.23**	0.83 (NS)
8/21	Between	2	253.68	2
	Within	21	104.3	21
	F =		2.43NS	0.09 (NS)
9/13	Between	2	928.8	2
	Within	21	114.4	21
	F =		8.1**	1.40 (NS)
10/14	Between	2	1132.9	2
	Within	21	1206.6	21
	F =		9.8**	2.10 (NS)

* 5% significance level

** 1% significance level

TABLE B-3 cont'd (1986)

Bonferroni paired t-tests for dates with significant ANOVA differences, 1986:

DATE	HORIZON	CON/ANT	CON/GND	ANT/GND
6/16	ORGANIC	NS(p>.05)	NS(p>.05)	NS(p>.05)
7/23	"	***(p<.001)	**(p<.01)	NS(p>.05)
9/13	"	NS(p>.05)	NS(p>.05)	**(p<.01)
10/14	"	*(p<.05)	NS(p>.05)	**(p<.01)

TABLE B-4. SOIL MOISTURE (mean %, w/w \pm SD, n = 8) 1987.

HORIZON	CONTROL SITE		ANTENNA SITE		GROUND SITE	
	ORGANIC	MINERAL	ORGANIC	MINERAL	ORGANIC	MINERAL
DATE						
6/16	21 \pm 5	10 \pm 2	30 \pm 10	7 \pm 1	25 \pm 7	11 \pm 2
7/21	30 \pm 8	18 \pm 2	35 \pm 8	15 \pm 3	31 \pm 7	19 \pm 3
8/25	31 \pm 8	15 \pm 2	34 \pm 11	11 \pm 4	32 \pm 7	15 \pm 2
9/9	34 \pm 7	14 \pm 3	31 \pm 6	10 \pm 2	33 \pm 7	14 \pm 3
10/16	35 \pm 10	16 \pm 1	29 \pm 4	12 \pm 1	42 \pm 7	16 \pm 1

ONE-WAY ANOVA

DATE	ORGANIC			MINERAL	
	D. F.	M. S.		D.F.	M.S.
6/16	Between	2	156.9	2	33.7
	Within	21	58.7	21	3.3
	F =		2.67 (NS)		10.24**
7/21	Between	2	3.5	2	40.4
	Within	21	50.6	21	7.7
	F =		0.07 (NS)		5.24*
8/25	Between	2	26.6	2	32.5
	Within	21	79.8	21	6.6
	F =		0.33 (NS)		4.94*
9/9	Between	2	24.5	2	38.5
	Within	21	46.3	21	6.1
	F =		0.53 (NS)		6.33**
10/16	Between	2	324.6	2	38.9
	Within	21	53.2	21	0.5
	F =		6.11**		76.40**

* 5% significance level

** 1% significance level

TABLE B-4 cont'd (1987)

Bonferroni paired t-tests for dates with significant ANOVA differences, 1987:

DATE	HORIZON	CON/ANT	CON/GND	ANT/GND
6/16	MINERAL	*(p<.05)	NS(p>.05)	**(p<.01)
7/21	"	NS(p>.05)	NS(p>.05)	*(p<.05)
8/25	"	*(p<.05)	NS(p>.05)	NS(p>.05)
9/9	"	*(p<.05)	NS(p>.05)	*(p<.05)
10/16	ORGANIC	NS(p>.05)	NS(p>.05)	*(p<.05)
10/16	MINERAL	***(p<.001)	NS(p>.05)	***(p<.001)

TABLE B-5. SOIL MOISTURE (mean %, w/w \pm SD, n = 8) 1988.

HORIZON	CONTROL SITE		ANTENNA SITE		GROUND SITE	
	ORGANIC	MINERAL	ORGANIC	MINERAL	ORGANIC	MINERAL
DATE						
6/13	25 \pm 4	9 \pm 2	24 \pm 8	10 \pm 1	22 \pm 7	9 \pm 1
7/8	25 \pm 9	12 \pm 2	22 \pm 5	12 \pm 4	22 \pm 5	9 \pm 2
8/22	24 \pm 8	10 \pm 1	28 \pm 7	8 \pm 2	24 \pm 8	8 \pm 2
9/11	38 \pm 6	13 \pm 2	40 \pm 5	13 \pm 1	40 \pm 3	14 \pm 3
10/11	36 \pm 5	16 \pm 2	34 \pm 6	14 \pm 2	33 \pm 6	16 \pm 2

ONE-WAY ANOVA: (between sites)

DATE	ORGANIC			MINERAL	
	D.F.	M.S.	F =	D.F.	M.S.
6/13	Between	2	12.5	2	0.2
	Within	21	42.8	21	1.5
			0.29 (NS)		0.13 (NS)
7/8	Between	2	25.6	2	26.2
	Within	21	47	21	7.7
			0.5 (NS)		3.40 (NS)
8/22	Between	2	28.2	2	4.8
	Within	21	60.2	21	2.8
			0.47 (NS)		2.77 (NS)
9/11	Between	2	11.6	2	1.5
	Within	21	22.7	21	4.5
			0.51 (NS)		0.33 (NS)
10/11	Between	2	6.7	2	14.5
	Within	21	26.3	21	5.4
			0.26 (NS)		2.69 (NS)

* 5% significance level

** 1% significance level

TABLE B-6. SOIL MOISTURE (mean %, w/w \pm SD, n = 8) 1989.

HORIZON	CONTROL SITE		ANTENNA SITE		GROUND SITE	
	ORG	MIN	ORG	MIN	ORG	MIN
DATE						
6/19	34 \pm 6	17 \pm 2	33 \pm 5	15 \pm 2	34 \pm 4	16 \pm 2
7/17	25 \pm 7	11 \pm 2	21 \pm 5	9 \pm 1	24 \pm 5	11 \pm 2
8/14	32 \pm 5	17 \pm 2	31 \pm 4	12 \pm 2	28 \pm 4	15 \pm 2
9/12	43 \pm 8	15 \pm 2	48 \pm 6	12 \pm 2	40 \pm 7	12 \pm 1
10/15	28 \pm 8	13 \pm 2	34 \pm 5	7 \pm 1	35 \pm 8	9 \pm 2

ONE-WAY ANOVA (between sites):

Date	ORGANIC			MINERAL	
	D.F.	M.S.	D.F.	M.S.	
6/19	Between	2	12.8	2	8.0
	Within	21	24.1	21	3.9
	F=		0.53 NS		2.09 NS
7/17	Between	2	28.67	2	13.2
	Within	21	32.08	21	2.6
	F=		0.89 NS		4.98 *
8/14	Between	2	28.59	2	40.5
	Within	21	18.06	21	3.7
	F=		1.52 NS		10.75 **
9/12	Between	2	123.4	2	28.6
	Within	21	49.2	21	2.2
	F=		2.50 NS		12.58 **
10/15	Between	2	114.6	2	74.4
	Within	21	48.3	21	2.6
	F=		2.37 NS		27.62 **

* = 5% significance level

**= 1% significance level

TABLE B-6 cont'd (1989)

Bonferroni paired t-tests for dates with significant ANOVA differences, 1989:

DATE	HORIZON	CON/ANT	CON/GND	ANT/GND
7/17	MINERAL	*(p<.05)	NS(p>.05)	NS(p>.05)
8/14	"	**(p<.01)	NS(p>.05)	NS(p>.05)
9/12	"	**(p<.01)	**(p<.01)	NS(p>.05)
10/15	"	***(p<.001)	**(p<.01)	**(p<.01)

TABLE B-7. SOIL MOISTURE (mean %, w/w \pm SD, n = 8) 1990.

HORIZON	CONTROL SITE		ANTENNA SITE		GROUND SITE	
	ORG	MIN	ORG	MIN	ORG	MIN
DATE						
6/19	31 \pm 4	14 \pm 2	32 \pm 3	17 \pm 4	35 \pm 6	18 \pm 1
7/16	32 \pm 9	12 \pm 1	27 \pm 5	15 \pm 8	30 \pm 8	16 \pm 1
8/14	23 \pm 4	8 \pm 1	26 \pm 4	14 \pm 3	25 \pm 5	14 \pm 2
9/17	49 \pm 8	16 \pm 5	44 \pm 9	6 \pm 3	38 \pm 4	13 \pm 2
10/14	53 \pm 11	17 \pm 5	43 \pm 8	19 \pm 2	33 \pm 2	18 \pm 2

ONE-WAY ANOVA (between sites):

Date	ORGANIC		MINERAL		
	D.F.	M.S.	D.F.	M.S.	
6/19	Between	2	29.31	2	30.19
	Within	21	19.05	21	6.57
	F=		1.54 NS		4.59 *
7/16	Between	2	50.43	2	43.39
	Within	21	54.97	21	22.55
	F=		0.92 NS		1.92 NS
8/14	Between	2	15.22	2	100.88
	Within	21	19.46	21	4.78
	F=		0.78 NS		21.1 **
9/17	Between	2	242.66	2	29.15
	Within	21	50.17	21	11.4
	F=		4.84 *		2.56 NS
10/14	Between	2	722.0	2	5.63
	Within	21	62.02	21	9.72
	F=		11.64 **		0.58 NS

* = 5% significance level

**= 1%/significance level

TABLE B-7 cont'd (1990)

Bonferroni paired t-tests for dates with significant ANOVA differences, 1990:

DATE	HORIZON	CON/ANT	CON/GND	ANT/GND
6/19	MINERAL	NS(p>.05)	*(p<.05)	NS(p>.05)
8/14	"	***(p<.001)	***(p<.001)	NS(p>.05)
9/17	ORGANIC	NS(p>.05)	*(p<.05)	NS(p>.05)
10/14	"	NS(p>.05)	***(p<.001)	NS(p>.05)

TABLE B-8. SOIL MOISTURE (mean %, w/w \pm SD, n = 8) 1991.

HORIZON	CONTROL SITE		ANTENNA SITE		GROUND SITE	
	ORGANIC	MINERAL	ORGANIC	MINERAL	ORGANIC	MINERAL
DATE						
6/10	31 \pm 5	17 \pm 2	31 \pm 5	14 \pm 3	18 \pm 8	14 \pm 3
7/15	27 \pm 4	15 \pm 2	33 \pm 9	14 \pm 4	29 \pm 8	15 \pm 2
8/12	24 \pm 6	13 \pm 2	21 \pm 6	8 \pm 2	20 \pm 6	10 \pm 3
9/24	41 \pm 9	14 \pm 2	45 \pm 8	15 \pm 3	32 \pm 5	17 \pm 3
10/20	40 \pm 6	15 \pm 3	40 \pm 6	12 \pm 1	37 \pm 6	15 \pm 2

ONE-WAY ANOVA (between sites)

DATE	ORGANIC		MINERAL		
	D. F.	M. S.	D.F.	M.S.	
6/10	Between	2	36.3	2	25.7
	Within	21	37.1	21	7.3
	F =		0.97		3.54*
7/15	Between	2	76	2	2.3
	Within	21	53.7	21	8.3
	F =		1.42		0.28
8/12	Between	2	21.4	2	51.1
	Within	21	36	21	5.7
	F =		0.59		9.01**
9/24	Between	2	371.8	2	20.3
	Within	21	62.1	21	6.7
	F =		5.99*		3.01
10/20	Between	2	22.7	2	23
	Within	21	35.3	21	4.5
	F =		0.64		5.09*

* 5% significance level

** 1% significance level

TABLE B-8 cont'd (1991)

Bonferroni paired t-tests for dates with significant ANOVA differences, 1991:

DATE	HORIZON	CON/ANT	CON/GND	ANT/GND
6/10	MINERAL	NS(p>.05)	NS(p>.05)	NS(p>.05)
8/12	"	**(p<.01)	*(p<.05)	NS(p>.05)
9/24	ORGANIC	NS(p>.05)	NS(p>.05)	*(p<.05)
10/20	MINERAL	NS(p>.05)	NS(p>.05)	*(p<.05)

TABLE B-9. SOIL MOISTURE (mean %, w/w \pm SD, n = 8) 1992.

ORGANIC HORIZON

	CONTROL SITE	ANTENNA SITE	GROUND SITE
DATE			
JUL 20	48 \pm 13	48 \pm 9	49 \pm 7
AUG 26	27 \pm 7	33 \pm 3	28 \pm 7
SEP 23	37 \pm 7	36 \pm 4	37 \pm 10

ONE-WAY ANOVA (between sites)

Date	ORGANIC		
	D.F.	M.S.	F
JUL20	Between	2	6.8
	Within	21	103.9
AUG26	Between	2	80.4
	Within	21	35.7
SEP23	Between	2	3.8
	Within	21	52.9

TABLE B-10. SOIL MOISTURE (mean %, w/w \pm SD, n = 8) 1993.

HORIZON	CONTROL SITE		ANTENNA SITE		GROUND SITE	
	ORGANIC	MINERAL	ORGANIC	MINERAL	ORGANIC	MINERAL
DATE						
6/16	42 \pm 10	15 \pm 3	38 \pm 8	12 \pm 4	41 \pm 3	13 \pm 3
7/21	44 \pm 11	14 \pm 2	36 \pm 7	8 \pm 1	39 \pm 9	12 \pm 3
8/15	48 \pm 9	16 \pm 3	39 \pm 11	13 \pm 4	36 \pm 5	17 \pm 2
9/16	50 \pm 8	9 \pm 2	47 \pm 9	17 \pm 2	44 \pm 10	18 \pm 2

ONE-WAY ANOVA (between sites)

DATE	ORGANIC			MINERAL	
	D.F.	M.S.	F =	D.F.	M.S.
6/6	Between	2	41.3	2	16.2
	Within	21	56.7	21	10.4
	F =		0.73		1.55
7/21	Between	2	129.0	2	55.0
	Within	21	85.7	21	4.5
	F =		1.50		12.2**
8/15	Between	2	266.8	2	29.6
	Within	21	74.5	21	9.7
	F =		3.57*		3.06
9/16	Between	2	84.9	2	1.8
	Within	21	82.6	21	3.8
	F =		1.03		0.47

* 5% significance level

** 1% significance level

TABLE B-10 cont'd (1993)

Bonferroni paired t-tests for dates with significant ANOVA differences, 1993:

DATE 7/21	HORIZON MINERAL	CON/ANT ***(p<.001)	CON/GND NS(p>.05)	ANT/GND *(p<.05)
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APPENDIX C

Amoeba Population Data

TABLE C-1. Density (mean \pm SE, n = 8) of total amoebae in each site and horizon, 1984-1992. Mean number/g soil expressed as $\log_{10} \pm$ SE and as an arithmetic value.

Total amoeba counts, 1984:

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/18	4.2360 \pm 0.0754	17,221
		6/27	3.4218 \pm 0.1482	2,642
		7/9	4.9306 \pm 0.1648	85,246
		7/18	4.9595 \pm 0.1971	91,099
		8/6	6.1951 \pm 0.1892	1,567,198
		9/3	3.6416 \pm 0.0874	4,382
		10/2	3.7919 \pm 0.0703	6,194
	Mineral	6/18	3.5216 \pm 0.1187	3,324
		6/27	3.2592 \pm 0.1309	1,817
		7/9	4.5872 \pm 0.0951	38,662
		7/18	4.8386 \pm 0.0813	68,975
		8/6	6.0445 \pm 0.1730	1,108,036
		9/3	3.2351 \pm 0.0282	1,718
		10/2	3.0968 \pm 0.0426	1,250
Antenna	Organic	6/18	4.1801 \pm 0.1619	15,142
		6/27	3.2260 \pm 0.0379	1,683
		7/9	4.7781 \pm 0.1196	60,006
		7/18	5.0645 \pm 0.1644	116,018
		8/6	6.3561 \pm 0.0946	2,270,476
		9/3	3.7416 \pm 0.0974	5,517
		10/2	3.5398 \pm 0.1417	3,466
	Mineral	6/18	3.3785 \pm 0.0912	2,391
		6/27	3.3610 \pm 0.1156	2,296
		7/9	4.4526 \pm 0.1411	28,353
		7/18	4.6801 \pm 0.1059	47,882
		8/6	6.1468 \pm 0.0436	1,402,266
		9/3	2.8608 \pm 0.4186	726
		10/2	3.2100 \pm 0.0685	1,622
Ground	Organic	6/18	4.0180 \pm 0.0937	10,424
		6/27	3.4932 \pm 0.0551	3,113
		7/9	4.9379 \pm 0.1334	86,682
		7/18	4.7029 \pm 0.1015	50,458
		8/6	6.3718 \pm 0.0546	2,354,408
		9/3	3.5375 \pm 0.0941	3,448
		10/2	3.5104 \pm 0.0891	3,239
	Mineral	6/18	3.6139 \pm 0.1514	4,111
		6/27	3.1617 \pm 0.0933	1,451
		7/9	4.7428 \pm 0.0655	55,321
		7/18	4.7081 \pm 0.0923	51,067
		8/6	6.0817 \pm 0.0249	1,207,183
		9/3	3.2883 \pm 0.0711	1,943
		10/2	3.0809 \pm 0.0311	1,205

TABLE C-1 cont'd (total amoeba counts, 1985)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/10	3.2624 ± 0.0652	1,830
		6/28	4.0844 ± 0.0839	12,149
		7/22	3.6077 ± 0.0812	4,053
		8/20	6.1766 ± 0.0643	1,502,118
		9/6	4.5358 ± 0.0634	34,340
		10/5	5.8135 ± 0.0812	650,000
	Mineral	6/10	2.9111 ± 0.0416	815
		6/28	3.2080 ± 0.0387	1,615
		7/22	3.0907 ± 0.0492	1,232
		8/20	5.8769 ± 0.0770	753,272
		9/6	4.3583 ± 0.0760	22,820
		10/5	5.5159 ± 0.1039	328,046
Antenna	Organic	6/10	3.4329 ± 0.1000	2,710
		6/28	4.2085 ± 0.0770	16,163
		7/22	3.6775 ± 0.0881	4,760
		8/20	6.0385 ± 0.1176	1,092,884
		9/6	4.6148 ± 0.1168	41,195
		10/5	5.6130 ± 0.1714	410,309
	Mineral	6/10	2.8932 ± 0.0607	782
		6/28	3.2826 ± 0.0334	1,917
		7/22	2.9822 ± 0.0509	960
		8/20	5.8234 ± 0.0708	666,007
		9/6	4.4207 ± 0.0371	26,348
		10/5	5.3618 ± 0.1225	230,051
Ground	Organic	6/10	3.4939 ± 0.1079	3,118
		6/28	4.0863 ± 0.1101	12,200
		7/22	3.7364 ± 0.0912	5,452
		8/20	6.1303 ± 0.1064	1,350,060
		9/6	4.5629 ± 0.0518	36,552
		10/5	5.6741 ± 0.0889	472,226
	Mineral	6/10	2.8187 ± 0.0275	659
		6/28	3.2690 ± 0.0352	1,858
		7/22	3.0474 ± 0.0487	1,115
		8/20	5.8654 ± 0.0600	733,598
		9/6	4.2669 ± 0.0673	18,488
		10/5	5.5074 ± 0.0730	321,703

TABLE C-1 cont'd (total amoeba counts, 1986)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/16	3.2351 ± 0.0719	1,868
		7/23	3.8417 ± 0.1143	8,712
		8/21	3.2858 ± 0.0709	2,119
		9/13	3.4383 ± 0.0418	2,840
		10/14	3.5474 ± 0.0418	3,650
	Mineral	6/16	2.9239 ± 0.0491	878
		7/23	3.3503 ± 0.0616	2,394
		8/21	3.1619 ± 0.0340	1,482
		9/13	3.0334 ± 0.0532	1,136
		10/14	3.0958 ± 0.0532	1,311
Antenna	Organic	6/16	3.1815 ± 0.0599	1,631
		7/23	4.1665 ± 0.1098	18,313
		8/21	3.4677 ± 0.0881	3,466
		9/13	3.4743 ± 0.0390	3,062
		10/14	3.4527 ± 0.0600	3,944
	Mineral	6/16	2.7746 ± 0.0291	605
		7/23	3.3356 ± 0.0764	2,410
		8/21	3.2013 ± 0.0502	1,667
		9/13	3.1857 ± 0.0546	1,613
		10/14	3.2539 ± 0.0502	1,882
Ground	Organic	6/16	3.0296 ± 0.0459	1,100
		7/23	3.9613 ± 0.1292	12,821
		8/21	3.3581 ± 0.0841	2,613
		9/13	3.3149 ± 0.0761	2,268
		10/14	3.4694 ± 0.0776	3,303
	Mineral	6/16	2.8546 ± 0.0584	763
		7/23	3.5216 ± 0.0763	3,698
		8/21	3.1756 ± 0.0619	1,610
		9/13	3.0774 ± 0.0612	1,271
		10/14	3.0908 ± 0.0526	1,288

TABLE C-1 cont'd (total amoeba counts, 1987)

SITE	HORIZON	Date	MEAN (LOG)	MEAN
Control	Organic	6/16	3.5023 ± 0.0667	3,454
		7/21	4.2649 ± 0.0674	19,990
		8/25	4.2584 ± 0.1098	22,599
		9/9	3.5606 ± 0.0677	3,934
		10/16	3.4122 ± 0.1183	3,345
	Mineral	6/16	3.2202 ± 0.0562	1,756
		7/21	4.0187 ± 0.0469	10,884
		8/25	4.0410 ± 0.1242	15,281
		9/9	2.9155 ± 0.0685	905
		10/16	2.7697 ± 0.0520	619
Antenna	Organic	6/16	3.9775 ± 0.0596	10,121
		7/21	4.6896 ± 0.0591	52,262
		8/25	4.5011 ± 0.0868	37,430
		9/9	3.3702 ± 0.0761	2,572
		10/16	3.3885 ± 0.1472	2,822
	Mineral	6/16	3.6720 ± 0.0430	4,869
		7/21	4.0610 ± 0.0613	10,622
		8/25	4.2035 ± 0.1089	19,225
		9/9	2.8870 ± 0.0457	803
		10/16	2.6688 ± 0.0385	481
Ground	Organic	6/16	3.7142 ± 0.0674	5,724
		7/21	4.2112 ± 0.1141	20,703
		8/25	4.5521 ± 0.0868	40,689
		9/9	3.5906 ± 0.0838	4,422
		10/16	3.1822 ± 0.0546	1,607
	Mineral	6/16	3.2228 ± 0.0469	1,720
		7/21	3.5745 ± 0.0280	3,811
		8/25	4.3155 ± 0.0579	22,048
		9/9	2.9739 ± 0.0689	1,020
		10/16	2.7394 ± 0.0263	556

TABLE C-1 cont'd (total amoeba counts, 1988)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/13	3.6012 ± 0.0599	4,595
		7/08	4.1868 ± 0.1343	20,409
		8/22	3.5027 ± 0.1571	5,375
		9/11	3.1352 ± 0.0419	1,408
		10/11	3.0067 ± 0.1105	1,439
	Mineral	6/13	2.7019 ± 0.0313	512
		7/08	3.6115 ± 0.0878	4,730
		8/22	3.0178 ± 0.0885	1,224
		9/11	2.6798 ± 0.0388	493
		10/11	2.8213 ± 0.0576	708
Antenna	Organic	6/13	3.5300 ± 0.0930	3,919
		7/08	3.8852 ± 0.1166	9,805
		8/22	3.7094 ± 0.1676	9,004
		9/11	3.0418 ± 0.0841	1,273
		10/11	2.8586 ± 0.1417	1,131
	Mineral	6/13	2.7117 ± 0.0449	534
		7/08	3.5734 ± 0.1008	4,459
		8/22	3.0705 ± 0.0463	1,228
		9/11	2.6560 ± 0.0258	459
		10/11	2.8555 ± 0.0555	762
Ground	Organic	6/13	3.5642 ± 0.0599	3,915
		7/08	4.1868 ± 0.1343	17,461
		8/22	3.7129 ± 0.1441	8,440
		9/11	3.0177 ± 0.0548	1,099
		10/11	2.9625 ± 0.0910	1,078
	Mineral	6/13	2.7178 ± 0.0350	534
		7/08	3.5449 ± 0.0663	3,815
		8/22	2.9993 ± 0.0451	1,037
		9/11	2.6976 ± 0.0372	512
		10/11	2.8321 ± 0.0502	715

TABLE C-1 cont'd (total amoeba counts, 1989)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/19	3.2510 ± 0.0574	1,896
		7/17	3.5023 ± 0.1174	4,235
		8/14	4.1548 ± 0.1816	31,842
		9/12	3.8811 ± 0.2058	15,325
		10/15	3.3560 ± 0.1017	2,979
	Mineral	6/19	2.8674 ± 0.0489	769
		7/17	3.3009 ± 0.0641	2,162
		8/14	3.4678 ± 0.1524	4,447
		9/12	2.9249 ± 0.0682	917
		10/15	2.8636 ± 0.1205	1,075
Antenna	Organic	6/19	3.2294 ± 0.0777	1,879
		7/17	4.2059 ± 0.1597	21,346
		8/14	4.0351 ± 0.1878	21,947
		9/12	3.4824 ± 0.1430	4,548
		10/15	3.6665 ± 0.1338	6,666
	Mineral	6/19	2.7368 ± 0.0528	577
		7/17	3.3662 ± 0.1278	3,157
		8/14	3.2072 ± 0.0622	1,735
		9/12	2.9012 ± 0.0977	952
		10/15	2.8436 ± 0.1205	1,169
Ground	Organic	6/19	3.1575 ± 0.0683	1,574
		7/17	3.6423 ± 0.1130	5,432
		8/14	4.1852 ± 0.0981	18,234
		9/12	3.5597 ± 0.1322	5,013
		10/15	3.7225 ± 0.1462	7,624
	Mineral	6/19	2.7656 ± 0.0461	607
		7/17	3.2489 ± 0.0729	1,928
		8/14	3.3163 ± 0.0572	2,194
		9/12	2.9759 ± 0.1046	1,188
		10/15	2.8110 ± 0.0687	709

TABLE C-1 cont'd (total amoeba counts, 1990)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/19	4.5119 ± 0.2888	32,501
		7/16	4.3348 ± 0.2441	68,593
		8/14	4.6732 ± 0.1320	67,483
		9/17	5.5479 ± 0.1286	449,366
		10/14	5.3436 ± 0.0555	233,526
	Mineral	6/19	3.6791 ± 0.1452	6,597
		7/16	3.9061 ± 0.1178	10,081
		8/14	4.6378 ± 0.0952	51,350
		9/17	4.8346 ± 0.0777	76,493
		10/14	4.8097 ± 0.2020	124,118
Antenna	Organic	6/19	4.1527 ± 0.1671	24,121
		7/16	4.7737 ± 0.1350	79,835
		8/14	4.9369 ± 0.1036	107,769
		9/17	5.1380 ± 0.0915	161,554
		10/14	5.6321 ± 0.1600	651,927
	Mineral	6/19	3.5838 ± 0.1593	5,574
		7/16	4.2308 ± 0.0716	18,623
		8/14	4.5975 ± 0.0967	48,736
		9/17	4.2191 ± 0.2449	27,756
		10/14	4.9791 ± 0.0753	106,446
Ground	Organic	6/19	3.8654 ± 0.1527	10,969
		7/16	4.5587 ± 0.0758	40,719
		8/14	4.8353 ± 0.1154	83,484
		9/17	4.9736 ± 0.0918	113,196
		10/14	5.3623 ± 0.0569	242,543
	Mineral	6/19	3.5524 ± 0.0828	4,027
		7/16	3.9625 ± 0.1462	12,915
		8/14	4.6473 ± 0.1492	63,241
		9/17	4.5940 ± 0.1569	57,468
		10/14	4.6801 ± 0.0665	51,737

TABLE C-1 cont'd(total amoeba counts, 1991)

33

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/10	3.7945 ± 0.1131	7,738
		7/15	3.8307 ± 0.0788	7,437
		8/12	3.8047 ± 0.0973	7,673
		9/24	4.8927 ± 0.0733	86,201
		10/20	4.1747 ± 0.0762	16,575
	Mineral	6/10	2.9686 ± 0.0389	958
		7/15	3.0634 ± 0.0776	1,303
		8/12	3.0943 ± 0.0651	1,344
		9/24	4.1524 ± 0.0801	15,862
		10/20	3.4738 ± 0.0794	3,330
Antenna	Organic	6/10	3.8747 ± 0.1564	10,436
		7/15	4.2185 ± 0.1080	20,055
		8/12	3.8350 ± 0.0685	7,552
		9/24	4.7986 ± 0.0876	72,341
		10/20	4.0353 ± 0.1395	15,067
	Mineral	6/10	2.9008 ± 0.0393	820
		7/15	2.8840 ± 0.0550	810
		8/12	3.0934 ± 0.0776	1,394
		9/24	4.0654 ± 0.0856	13,396
		10/20	3.2778 ± 0.0591	2,028
Ground	Organic	6/10	3.6119 ± 0.1514	6,687
		7/15	4.0755 ± 0.1252	16,985
		8/12	3.7108 ± 0.0775	5,784
		9/24	4.6986 ± 0.0852	56,946
		10/20	4.2019 ± 0.0950	18,682
	Mineral	6/10	2.8689 ± 0.0903	881
		7/15	3.0067 ± 0.0689	1,116
		8/12	2.9787 ± 0.0694	1,052
		9/24	3.9547 ± 0.0425	9,348
		10/20	3.2605 ± 0.0893	2,166

TABLE C-1 cont'd(total amoeba counts, 1992)

34

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	7/20	4.7251 \pm 0.0855	61,851
		8/26	3.8665 \pm 0.0284	7,471
		9/23	4.0538 \pm 0.0608	12,095
Antenna	Organic	7/20	4.5317 \pm 0.1716	58,538
		8/26	4.0124 \pm 0.0622	11,233
		9/23	4.0847 \pm 0.0878	13,799
Ground	Organic	7/20	5.0294 \pm 0.1442	168,600
		8/26	3.9838 \pm 0.0407	9,550
		9/23	3.9872 \pm 0.0474	9,839

TABLE C-2. Density (mean \pm SE, n = 8) of cysts in each site and horizon, 1984-1992. Mean number/g soil expressed as $\log_{10} \pm$ SE and as an arithmetic value.

Cyst counts, 1984:

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/18	3.0307 \pm 0.0599	1,073
		6/27	3.4173 \pm 0.0747	2,614
		7/9	3.3463 \pm 0.0991	2,220
		7/18	5.2551 \pm 0.0624	179,908
		8/6	5.0596 \pm 0.1761	114,714
		9/3	3.1713 \pm 0.0603	1,483
		10/2	3.4553 \pm 0.0312	2,853
	Mineral	6/18	3.5558 \pm 0.1488	3,596
		6/27	3.3614 \pm 0.1177	2,298
		7/9	3.0666 \pm 0.0297	1,166
		7/18	5.1182 \pm 0.0992	131,308
		8/6	4.6310 \pm 0.0961	42,766
		9/3	2.7118 \pm 0.0320	515
		10/2	3.1109 \pm 0.0596	1,291
Antenna	Organic	6/18	3.4091 \pm 0.1188	2,565
		6/27	3.5337 \pm 0.0927	3,418
		7/9	3.1367 \pm 0.0582	1,370
		7/18	4.4346 \pm 0.1529	27,203
		8/6	5.0093 \pm 0.1776	102,171
		9/3	2.9474 \pm 0.0628	886
		10/2	3.4906 \pm 0.0958	3,095
	Mineral	6/18	3.2737 \pm 0.1258	1,878
		6/27	3.2668 \pm 0.0599	1,849
		7/9	2.9683 \pm 0.0360	929
		7/18	4.4752 \pm 0.1067	29,873
		8/6	4.7276 \pm 0.0629	53,402
		9/3	2.7293 \pm 0.0328	536
		10/2	3.2941 \pm 0.0732	1,898
Ground	Organic	6/18	3.2699 \pm 0.0834	1,862
		6/27	3.3131 \pm 0.1147	2,056
		7/9	3.5715 \pm 0.0646	3,729
		7/18	3.9097 \pm 0.2274	8,124
		8/6	5.1981 \pm 0.0817	157,818
		9/3	2.9114 \pm 0.0524	815
		10/2	3.4532 \pm 0.0759	2,840
	Mineral	6/18	3.2361 \pm 0.1548	1,722
		6/27	3.0597 \pm 0.0439	1,147
		7/9	2.9218 \pm 0.0429	835
		7/18	4.1143 \pm 0.0950	13,013
		8/6	4.8872 \pm 0.0495	77,134
		9/3	2.7258 \pm 0.0599	532
		10/2	3.0192 \pm 0.0385	1,045

TABLE C-2 cont'd (Cyst counts, 1985)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/10	3.2885 ± 0.0904	1,944
		6/28	3.1028 ± 0.0286	1,267
		7/22	3.3157 ± 0.0624	2,069
		8/20	4.2243 ± 0.0462	16,764
		9/6	3.7867 ± 0.0670	6,119
		10/5	3.3753 ± 0.0749	2,373
	Mineral	6/10	2.9115 ± 0.0697	816
		6/28	3.0600 ± 0.0548	1,148
		7/22	2.6577 ± 0.0331	455
		8/20	3.4934 ± 0.0567	3,115
		9/6	2.8471 ± 0.0651	703
		10/5	1.9478 ± 0.0302	887
Antenna	Organic	6/10	3.3412 ± 0.0998	2,194
		6/28	3.8085 ± 0.0813	6,434
		7/22	3.4936 ± 0.0484	3,117
		8/20	4.2677 ± 0.0952	18,526
		9/6	3.7528 ± 0.0401	5,661
		10/5	3.5396 ± 0.0956	3,465
	Mineral	6/10	2.6201 ± 0.0138	417
		6/28	3.1461 ± 0.0428	1,400
		7/22	2.7350 ± 0.0874	543
		8/20	3.4618 ± 0.0706	2,896
		9/6	3.2377 ± 0.1090	1,729
		10/5	3.1154 ± 0.0504	1,305
Ground	Organic	6/10	3.2886 ± 0.0723	1,851
		6/28	3.8052 ± 0.0551	6,386
		7/22	3.3645 ± 0.0768	2,315
		8/20	4.2313 ± 0.0557	17,037
		9/6	3.2807 ± 0.1658	1,909
		10/5	3.5679 ± 0.0722	3,698
	Mineral	6/10	2.7290 ± 0.0396	536
		6/28	2.8715 ± 0.0387	744
		7/22	2.6475 ± 0.0247	444
		8/20	3.4148 ± 0.0347	2,599
		9/6	3.0578 ± 0.0875	1,142
		10/5	3.1878 ± 0.0299	1,541

TABLE C-2 cont'd (Cyst counts, 1986)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/16	3.1229 ± 0.1022	1,487
		7/23	3.1633 ± 0.0704	1,627
		8/21	3.0034 ± 0.0587	1,085
		9/13	3.1226 ± 0.0532	1,405
		10/14	3.2315 ± 0.0531	1,905
	Mineral	6/16	2.7723 ± 0.0388	610
		7/23	2.8795 ± 0.1737	762
		8/21	2.8795 ± 0.0307	771
		9/13	2.7041 ± 0.0324	517
		10/14	2.7665 ± 0.0324	597
Antenna	Organic	6/16	3.0027 ± 0.0316	1,027
		7/23	3.1246 ± 0.0564	1,410
		8/21	3.0394 ± 0.0347	1,121
		9/13	3.3280 ± 0.0570	1,978
		10/14	3.0955 ± 0.0347	1,275
	Mineral	6/16	2.7324 ± 0.0325	552
		7/23	2.8698 ± 0.0282	753
		8/21	2.7540 ± 0.0120	569
		9/13	2.7098 ± 0.0284	521
		10/14	2.8068 ± 0.0120	643
Ground	Organic	6/16	2.9317 ± 0.0272	866
		7/23	3.3293 ± 0.0769	2,391
		8/21	3.0568 ± 0.0578	1,222
		9/13	3.0481 ± 0.0444	935
		10/14	3.2436 ± 0.0444	1,817
	Mineral	6/16	2.6945 ± 0.0349	507
		7/23	2.8955 ± 0.0387	786
		8/21	2.8178 ± 0.0327	671
		9/13	2.7807 ± 0.0514	642
		10/14	2.7700 ± 0.0106	590

TABLE C-2 cont'd (Cyst counts, 1987)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/16	3.4184 ± 0.0938	3,216
		7/21	4.2483 ± 0.0950	20,817
		8/25	3.8903 ± 0.0828	8,732
		9/9	3.4463 ± 0.0695	3,041
		10/16	3.5452 ± 0.0951	3,614
	Mineral	6/16	2.9241 ± 0.0371	863
		7/21	3.6005 ± 0.0379	4,091
		8/25	3.0145 ± 0.0764	1,156
		9/9	2.9036 ± 0.0488	807
		10/16	2.8454 ± 0.0520	738
Antenna	Organic	6/16	3.6068 ± 0.0617	4,292
		7/21	4.2492 ± 0.0458	18,526
		8/25	3.8237 ± 0.1268	8,917
		9/9	3.5535 ± 0.0670	3,883
		10/16	3.5104 ± 0.0747	3,539
	Mineral	6/16	2.8739 ± 0.0301	761
		7/21	3.5301 ± 0.0866	3,828
		8/25	3.0590 ± 0.0645	1,243
		9/9	2.9635 ± 0.0565	976
		10/16	2.6216 ± 0.0246	423
Ground	Organic	6/16	3.6124 ± 0.0951	4,564
		7/21	4.2294 ± 0.0598	18,026
		8/25	4.0436 ± 0.1150	13,892
		9/9	3.5258 ± 0.1054	4,116
		10/16	3.5002 ± 0.0829	3,118
	Mineral	6/16	2.8459 ± 0.0419	723
		7/21	3.6642 ± 0.0890	5,286
		8/25	3.0974 ± 0.0768	1,391
		9/9	2.9503 ± 0.0706	976
		10/16	2.8170 ± 0.0283	666

TABLE C-2 cont'd(cyst counts, 1988)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/13	3.2014 ± 0.0438	1,652
		7/08	3.2811 ± 0.0701	2,115
		8/22	3.2570 ± 0.0664	1,981
		9/11	3.3826 ± 0.0537	2,563
		10/11	2.8877 ± 0.0707	869
	Mineral	6/13	2.6892 ± 0.0301	497
		7/08	2.7125 ± 0.0227	520
		8/22	2.7826 ± 0.0488	632
		9/11	2.9108 ± 0.0799	926
		10/11	2.6999 ± 0.0245	507
Antenna	Organic	6/13	3.1759 ± 0.0666	1,649
		7/08	3.0890 ± 0.0315	1,250
		8/22	3.2463 ± 0.0421	1,821
		9/11	3.2449 ± 0.0588	1,879
		10/11	2.9356 ± 0.0537	910
	Mineral	6/13	2.6625 ± 0.0281	487
		7/08	2.6891 ± 0.0293	497
		8/22	2.7963 ± 0.0488	657
		9/11	2.9438 ± 0.0532	926
		10/11	2.6480 ± 0.0178	447
Ground	Organic	6/13	3.2162 ± 0.0498	1,721
		7/08	3.2353 ± 0.0735	1,902
		8/22	3.0631 ± 0.0409	1,192
		9/11	3.1970 ± 0.0588	1,694
		10/11	2.9220 ± 0.0547	884
	Mineral	6/13	2.6427 ± 0.0273	445
		7/08	2.6542 ± 0.0358	462
		8/22	2.8268 ± 0.0479	700
		9/11	2.9156 ± 0.0487	862
		10/11	2.6870 ± 0.0350	498

TABLE C-2 cont'd (cyst counts, 1989)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/19	3.0135 ± 0.0465	1,085
		7/17	2.8622 ± 0.0263	738
		8/14	3.6858 ± 0.1001	5,759
		9/12	3.4446 ± 0.1993	5,395
		10/15	3.1480 ± 0.0897	1,612
	Mineral	6/19	2.6719 ± 0.0125	472
		7/17	2.7453 ± 0.0417	575
		8/14	3.0393 ± 0.0582	1,163
		9/12	2.9111 ± 0.0621	876
		10/15	2.8352 ± 0.1271	1,077
Antenna	Organic	6/19	3.0407 ± 0.0432	1,150
		7/17	3.2068 ± 0.0785	1,793
		8/14	4.2982 ± 0.1158	24,695
		9/12	3.1153 ± 0.0498	1,368
		10/15	3.2187 ± 0.1022	1,958
	Mineral	6/19	2.6570 ± 0.0164	457
		7/17	2.9240 ± 0.0764	936
		8/14	3.2311 ± 0.0373	1,751
		9/12	2.8494 ± 0.0683	778
		10/15	2.9428 ± 0.1345	1,374
Ground	Organic	6/19	3.0350 ± 0.0477	1,144
		7/17	3.5060 ± 0.0728	3,497
		8/14	3.7366 ± 0.1249	7,236
		9/12	3.2394 ± 0.1004	2,060
		10/15	3.1688 ± 0.0941	1,737
	Mineral	6/19	2.6474 ± 0.0311	456
		7/17	2.8160 ± 0.0449	679
		8/14	3.3865 ± 0.0678	2,626
		9/12	2.7738 ± 0.3810	611
		10/15	2.7399 ± 0.0978	710

TABLE C-2 cont'd (cyst counts, 1990)

SITE	HORIZON	DATE	MEAN (LOG)	MEAN
Control	Organic	6/19	3.8695 ± 0.1179	9,364
		7/16	4.7692 ± 0.1047	74,428
		8/14	3.9928 ± 0.0809	11,102
		9/17	5.5601 ± 0.0670	391,725
		10/14	4.2924 ± 0.0972	23,056
	Mineral	6/19	3.5718 ± 0.1305	5,007
		7/16	4.0150 ± 0.1433	17,817
		8/14	3.8390 ± 0.1439	10,130
		9/17	4.5204 ± 0.0638	35,776
		10/14	3.3454 ± 0.0642	2,399
Antenna	Organic	6/19	3.3358 ± 0.0765	2,402
		7/16	4.5693 ± 0.1979	74,597
		8/14	4.6546 ± 0.0800	49,724
		9/17	4.7147 ± 0.1310	72,677
		10/14	4.7690 ± 0.2613	117,029
	Mineral	6/19	3.5747 ± 0.1227	5,072
		7/16	3.7022 ± 0.1785	11,940
		8/14	4.0618 ± 0.2124	35,706
		9/17	4.2752 ± 0.1166	24,979
		10/14	3.4814 ± 0.0641	3,289
Ground	Organic	6/19	3.6389 ± 0.1466	6,729
		7/16	4.9733 ± 0.1166	116,044
		8/14	4.4765 ± 0.1507	42,623
		9/17	5.0615 ± 0.1154	143,452
		10/14	4.5067 ± 0.1623	55,065
	Mineral	6/19	3.7880 ± 0.1308	7,884
		7/16	4.1989 ± 0.1237	22,177
		8/14	3.7018 ± 0.1332	6,771
		9/17	4.2890 ± 0.1839	31,020
		10/14	3.5688 ± 0.1094	4,567

TABLE C-2 cont'd(cyst counts, 1991)

SITE	HORIZON	DATE	MEAN	MEAN
Control	Organic	6/10	3.8144 \pm 0.1306	8,633
		7/15	3.6851 \pm 0.0978	5,707
		8/12	3.6919 \pm 0.1155	6,260
		9/24	4.5114 \pm 0.1217	43,867
		10/20	4.2275 \pm 0.1468	24,631
	Mineral	6/10	2.8505 \pm 0.0192	714
		7/15	2.8181 \pm 0.0541	703
		8/12	3.0374 \pm 0.0635	1,182
		9/24	3.8124 \pm 0.1308	9,387
		10/20	3.4550 \pm 0.1068	3,517
Antenna	Organic	6/10	3.9679 \pm 0.1637	14,264
		7/15	3.8121 \pm 0.0957	7,864
		8/12	4.1762 \pm 0.1066	18,197
		9/24	4.6781 \pm 0.1183	62,500
		10/20	3.8797 \pm 0.1401	11,091
	Mineral	6/10	2.9998 \pm 0.0509	1,049
		7/15	2.8558 \pm 0.0655	786
		8/12	3.1458 \pm 0.0458	1,449
		9/24	4.3704 \pm 0.1690	36,859
		10/20	3.2638 \pm 0.0606	1,980
Ground	Organic	6/10	3.8762 \pm 0.1471	12,294
		7/15	4.1659 \pm 0.1130	17,674
		8/12	3.8472 \pm 0.0838	7,917
		9/24	4.7363 \pm 0.1006	63,764
		10/20	3.7601 \pm 0.1126	7,162
	Mineral	6/10	2.9044 \pm 0.0425	830
		7/15	3.1858 \pm 0.0883	1,772
		8/12	3.0873 \pm 0.0593	1,300
		9/24	3.7331 \pm 0.0845	6,094
		10/20	3.6988 \pm 0.1404	6,845

TABLE C-2 cont'd (cyst counts, 1992)

SITE	HORIZON	DATE	MEAN	MEAN
Control	Organic	7/20	4.4462 ± 0.1591	44,103
		8/26	3.5328 ± 0.0491	3,570
		9/23	3.7050 ± 0.0439	5,250
Antenna	Organic	7/20	4.1944 ± 0.0794	17,992
		8/26	3.5038 ± 0.0348	3,261
		9/23	3.6648 ± 0.0347	4,732
Ground	Organic	7/20	4.6858 ± 0.1040	60,020
		8/26	3.5603 ± 0.0511	3,428
		9/23	3.6934 ± 0.0399	4,724

TABLE C-3. One-way analysis of variance by date and horizon, data log transformed. Significant differences at the 5% level (*) and the 1% level (**) indicated, paired t-tests between sites given, NS indicates no significant difference between sites for a given site and horizon.

TOTAL COUNT					
HORIZON	DATE	GROUPS	DF	MS	F
ORGANIC	6/18	among	2	0.1026	
		within	21	0.1085	0.9455 NS
	6/27	among	2	0.1531	
		within	21	0.0705	2.1719 NS
	7/9	among	2	0.0652	
		within	21	0.1580	0.4128 NS
	7/18	among	2	0.2768	
		within	21	0.2031	1.3627 NS
	8/6	among	2	0.0765	
		within	21	0.0127	0.6014 NS
MINERAL	9/3	among	2	0.0833	
		within	21	0.0693	1.2023 NS
	10/2	among	2	0.1915	
		within	21	0.0879	2.1793 NS
	6/18	among	2	0.1125	
		within	21	0.1208	0.9311 NS
	6/27	among	2	0.0794	
		within	21	0.1045	0.7597 NS
	7/9	among	2	0.1687	
		within	21	0.0886	1.9034 NS
	7/18	among	2	0.0572	
		within	21	0.0703	0.8146 NS
	8/6	among	2	0.0215	
		within	21	0.0865	0.7822 NS
	9/3	among	2	0.4341	
		within	21	0.4829	0.8989 NS
	10/2	among	2	0.0397	
		within	21	0.0199	1.9894 NS

TABLE C-3 cont'd(ANOVA 1984)

CYST COUNT					
HORIZON	DATE	GROUPS	DF	MS	F
ORGANIC	6/18	among	2	0.2930	
		within	21	0.0655	4.4677 **
	6/27	among	2	0.0975	
		within	21	0.0728	1.3379 NS
	7/9	among	2	0.3783	
		within	21	0.0464	8.1591 **
	7/18	among	2	3.6784	
		within	21	0.2106	17.4614 **
	8/6	among	2	0.0765	
		within	21	0.1845	0.4149 NS
	9/3	among	2	0.1589	
		within	21	0.0274	5.7869 *
MINERAL	10/2	among	2	0.0035	
		within	21	0.0424	0.0831 NS
MINERAL	6/18	among	2	0.2442	
		within	21	0.1652	1.4780 NS
	6/27	among	2	0.1905	
		within	21	0.0516	3.6859 *
	7/9	among	2	0.0437	
		within	21	0.0107	4.0797 *
	7/18	among	2	2.0687	
		within	21	0.0807	25.6121 **
	8/6	among	2	0.1339	
		within	21	0.0417	3.2041 NS
	9/3	among	2	0.0007	
		within	21	0.0152	0.0462 NS
MINERAL	10/2	among	2	0.1566	
		within	21	0.0277	5.6517 *

Bonferroni paired t-tests for dates with significant ANOVA differences, 1984:

DATE	HORIZON	COUNT	P-VALUES		
			CON/ANT	CON/GND	ANT/GND
6/18	ORGANIC	CYST	*(p<.05)	NS(p>.05)	NS(p>.05)
7/9	"	"	NS(p>.05)	NS(p>.05)	**(p<.01)
7/18	"	"	**(p<.01)	***(p<.001)	NS(p>.05)
9/3	"	"	NS(p>.05)	*(p<.05)	NS(p>.05)
6/27	MINERAL	"	NS(p>.05)	NS(p>.05)	NS(p>.05)
7/9	"	"	NS(p>.05)	*(p<.05)	NS(p>.05)
7/18	"	"	**(p<.01)	***(p<.001)	NS(p>.05)
10/2	"	"	NS(p>.05)	NS(p>.05)	*(p<.05)

TABLE C-3 cont'd(ANOVA, 1985)

HORIZON	DATE	GROUPS	TOTAL	COUNT	F
			DF	MS	
ORGANIC	6/10	among	2	0.1151	
		within	21	0.0691	1.6675 NS
	6/28	among	2	0.0120	
		within	21	0.0669	0.5996 NS
	7/22	among	2	0.0323	
		within	21	0.0605	0.5494 NS
	8/20	among	2	0.0395	
		within	21	0.0781	0.5057 NS
	9/6	among	2	0.0129	
		within	21	0.0543	0.2375 NS
	10/5	among	2	0.0845	
		within	21	0.1169	0.7222 NS
MINERAL	6/10	among	2	0.0192	
		within	21	0.0165	1.1672 NS
	6/28	among	2	0.0126	
		within	21	0.0103	1.2240 NS
	7/22	among	2	0.0238	
		within	21	0.0195	1.2220 NS
	8/20	among	2	0.0632	
		within	21	0.0388	0.1628 NS
	9/6	among	2	0.0478	
		within	21	0.0312	1.5361 NS
	10/5	among	2	0.0601	
		within	21	0.0809	0.7416 NS
CYST COUNT					
ORGANIC	6/10	among	2	0.0074	
		within	21	0.0623	0.1182 NS
	6/28	among	2	1.3215	
		within	21	0.0278	47.4044 **
	7/22	among	2	0.0680	
		within	21	0.0324	0.1471 NS
	8/20	among	2	0.0437	
		within	21	0.0379	0.1151 NS
	9/6	among	2	0.6419	
		within	21	0.0895	7.1687 **
	10/5	among	2	0.0868	
		within	21	0.0532	1.6296 NS
MINERAL	6/10	among	2	0.1735	
		within	21	0.0176	9.8358 **
	6/28	among	2	0.1577	
		within	21	0.1689	9.3409 **
	7/22	among	2	0.1831	
		within	21	0.0249	0.7347 NS
	8/20	among	2	0.0125	
		within	21	0.0251	0.4986 NS
	9/6	among	2	0.3059	
		within	21	0.0635	4.8191 *
	10/5	among	2	0.1213	
		within	21	0.0116	10.4572 **

TABLE C-3 cont'd(1985)

Bonferroni paired t-tests for dates with significant ANOVA differences, 1985:

DATE	HORIZON	COUNT	P-VALUES		
			CON/ANT	CON/GND	ANT/GND
6/28	ORGANIC	CYST	***(p<.001)	***(p<.001)	NS(p>.05)
9/6	"	"	NS(p>.05)	*(p<.05)	*(p<.05)
6/10	MINERAL	"	**(p<.01)	*(p<.05)	NS(p>.05)
6/28	"	"	NS(p>.05)	*(p<.05)	**(p<.01)
9/6	"	"	*(p<.05)	NS(p>.05)	NS(p>.05)
10/5	"	"	*(p<.05)	**(p<.01)	NS(p>.05)

TABLE C-3 cont'd (ANOVA, 1986)

HORIZON	DATE	GROUPS	TOTAL	COUNT	F
			DF	MS	
ORGANIC	6/16	among	2	0.0854	
		within	21	0.0290	2.9444 NS
	7/23	among	2	0.2154	
		within	21	0.1109	1.9429 NS
	8/21	among	2	0.0671	
		within	21	0.0528	1.2692 NS
	9/13	among	2	0.0557	
		within	21	0.0241	2.3082 NS
	10/14	among	2	0.0203	
		within	21	0.0301	0.6759 NS
MINERAL	6/16	among	2	0.0444	
		within	21	0.0178	2.5011 NS
	7/23	among	2	0.0859	
		within	21	0.0411	2.0904 NS
	8/21	among	2	0.0032	
		within	21	0.0199	0.1609 NS
	9/13	among	2	0.0491	
		within	21	0.0254	1.9366 NS
	10/14	among	2	0.0688	
		within	21	0.0216	3.1875 NS
CYST COUNT					
ORGANIC	6/16	among	2	0.0746	
		within	21	0.0313	2.3856 NS
	7/23	among	2	0.0947	
		within	21	0.0375	2.5222 NS
	8/21	among	2	0.0059	
		within	21	0.0213	0.2768 NS
	9/13	among	2	0.1682	
		within	21	0.0215	7.8353 **
	10/14	among	2	0.0536	
		within	21	0.0158	3.3803 NS
MINERAL	6/16	among	2	0.0141	
		within	21	0.0099	1.1413 NS
	7/23	among	2	0.0035	
		within	21	0.0866	0.0156 NS
	8/21	among	2	0.0315	
		within	21	0.0058	5.4504 *
	9/13	among	2	0.0143	
		within	21	0.0183	1.2100 NS
	10/14	among	2	0.0039	
		within	21	0.0041	1.1680 NS

Bonferroni paired t-tests for dates with significant ANOVA differences, 1986:

DATE	HORIZON	COUNT	P-VALUES		
			CON/ANT	CON/GND	ANT/GND
9/13	ORGANIC	CYST	*(p<.05)	NS(p>.05)	**(p<.01)
8/21	MINERAL	"	*(p<.05)	NS(p>.05)	NS(p>.05)

TABLE C-3 cont'd (ANOVA, 1987)

HORIZON	DATE	GROUPS	TOTAL	COUNT	F
			DF	MS	
ORGANIC	6/16	among	2	0.4539	
		within	21	0.0335	13.5708 **
	7/21	among	2	0.5498	
		within	21	0.0561	9.8078 **
	8/25	among	2	0.1971	
		within	21	0.0724	2.7207 NS
	9/9	among	2	0.1139	
		within	21	0.0464	2.4567 NS
	10/16	among	2	0.1281	
		within	21	0.1025	1.2495 NS
MINERAL	6/16	among	2	0.5376	
		within	21	0.0189	28.3907 **
	7/21	among	2	0.5813	
		within	21	0.0179	32.5106 **
	8/25	among	2	0.1524	
		within	21	0.0816	1.8683 NS
	9/9	among	2	0.0157	
		within	21	0.0306	0.5145 NS
	10/16	among	2	0.0214	
		within	21	0.0130	1.6470 NS
CYST COUNT					
ORGANIC	6/16	among	2	0.0975	
		within	21	0.0578	1.6871 NS
	7/21	among	2	0.0011	
		within	21	0.0920	0.0262 NS
	8/25	among	2	0.1018	
		within	21	0.0964	1.0553 NS
	9/9	among	2	0.0247	
		within	21	0.0545	0.4535 NS
	10/16	among	2	0.0044	
		within	21	0.0574	0.0773 NS
MINERAL	6/16	among	2	0.0125	
		within	21	0.0108	1.1606 NS
	7/21	among	2	0.0359	
		within	21	0.0449	0.7989 NS
	8/25	among	2	0.0136	
		within	21	0.0424	0.3209 NS
	9/9	among	2	0.0079	
		within	21	0.0282	0.7584 NS
	10/16	among	2	0.1247	
		within	21	0.0109	11.4238 **

Bonferroni paired t-tests for dates with significant ANOVA differences, 1987:

P-VALUES

DATE	HORIZON	COUNT	CON/ANT	CON/GND	ANT/GND
6/16	ORGANIC	TOTAL	***(p<.001)	NS(p>.05)	*(p<.05)
7/21	"	"	**(p<.01)	NS(p>.05)	**(p<.01)
6/16	MINERAL	"	***(p<.001)	NS(p>.05)	***(p<.001)
7/21	"	"	NS(p>.05)	***(p<.001)	***(p<.001)
10/16	MINERAL	CYST	**(p<.01)	NS(p>.05)	**(p<.01)

TABLE C-3 cont'd (ANOVA, 1988)

HORIZON	DATE	GROUPS	TOTAL COUNT		F
			DF	MS	
ORGANIC	6/13	among	2	0.00888	
		within	21	0.05431	0.1636NS
	7/08	among	2	0.23673	
		within	21	0.10552	2.2434NS
	8/22	among	2	0.11593	
		within	21	0.19607	0.5913NS
	9/11	among	2	0.03083	
		within	21	0.03146	0.9802NS
	10/11	among	2	0.08676	
		within	21	0.10819	0.802 NS
MINERAL	6/13	among	2	0.00052	
		within	21	0.01127	0.458 NS
	7/08	among	2	0.00894	
		within	21	0.05936	0.1505NS
	8/22	among	2	0.01093	
		within	21	0.03202	0.3413NS
	9/11	among	2	0.00348	
		within	21	0.00947	0.3678NS
	10/11	among	2	0.00245	
		within	21	0.0238	0.1029NS
CYST COUNT					
ORGANIC	6/13	among	2	0.00332	
		within	21	0.02356	0.1408NS
	7/08	among	2	0.08054	
		within	21	0.03018	2.6687NS
	8/22	among	2	0.09503	
		within	21	0.02096	4.5348 *
	9/11	among	2	0.07428	
		within	21	0.02611	2.8448NS
	10/11	among	2	0.00486	
		within	21	0.02898	0.1678NS
MINERAL	6/13	among	2	0.00434	
		within	21	0.00653	0.6649NS
	7/08	among	2	0.00687	
		within	21	0.00699	0.9828NS
	8/22	among	2	0.0041	
		within	21	0.01924	0.213 NS
	9/11	among	2	0.00253	
		within	21	0.03088	0.082 NS
	10/11	among	2	0.00583	
		within	21	0.00571	1.0213NS

Bonferroni paired t-tests for dates with significant ANOVA differences, 1988:

DATE	HORIZON	COUNT	P-VALUES		
			CON/ANT NS(p>.05)	CON/GND NS(p>.05)	ANT/GND NS(p>.05)
8/22	ORGANIC	CYST			

TABLE C-3 cont'd(ANOVA, 1989)

HORIZON	DATE	GROUPS	TOTAL COUNT		F
			DF	MS	
ORGANIC	6/19	among	2	0.0191	
		within	21	0.0373	0.5129 NS
	7/17	among	2	1.1098	
		within	21	0.1388	7.9962 **
	8/14	among	2	0.0504	
		within	21	0.2077	0.2426 NS
	9/12	among	2	0.3577	
		within	21	0.2141	1.6702 NS
	10/15	among	2	0.3118	
		within	21	0.1323	0.1193 NS
MINERAL	6/19	among	2	0.0377	
		within	21	0.0195	1.9351 NS
	7/17	among	2	0.0277	
		within	21	0.0687	0.4027 NS
	8/14	among	2	0.1371	
		within	21	0.0810	1.6928 NS
	9/12	among	2	0.0117	
		within	21	0.0670	0.1743 NS
	10/15	among	2	0.0056	
		within	21	0.1077	0.9491 NS
CYST COUNT					
ORGANIC	6/19	among	2	0.0019	
		within	21	0.0189	0.0979 NS
	7/17	among	2	0.8303	
		within	21	0.0324	25.6201 **
	8/14	among	2	0.9241	
		within	21	0.1041	8.8761 **
	9/12	among	2	0.2212	
		within	21	0.1394	1.5864 NS
	10/15	among	2	0.0105	
		within	21	0.0729	0.1446 NS
MINERAL	6/19	among	2	0.0014	
		within	21	0.0042	0.3263 NS
	7/17	among	2	0.0648	
		within	21	0.0256	2.5338 NS
	8/14	among	2	0.2421	
		within	21	0.0250	9.6847 **
	9/12	among	2	0.0379	
		within	21	0.0266	1.4233 NS
	10/15	among	2	0.0824	
		within	21	0.1168	0.7056 NS

Bonferroni paired t-tests for dates with significant ANOVA differences, 1989:

DATE	HORIZON	COUNT	P-VALUES		
			CON/ANT	CON/GND	ANT/GND
7/17	ORGANIC	TOTAL	**(p<.01)	NS(p>.05)	*(p<.05)
7/17	"	CYST	**(p<.01)	***(p<.001)	*(p<.05)
8/14	"	"	**(p<.01)	NS(p>.05)	*(p<.05)
8/14	MINERAL	CYST	NS(p>.05)	**(p<.01)	NS(p>.05)

TABLE C-3 cont'd (ANOVA, 1990)

HORIZON	DATE	GROUPS	TOTAL COUNT		F
			DF	MS	
ORGANIC	6/19	among	2	0.8393	
		within	21	0.3591	2.3374 NS
	7/16	among	2	0.3854	
		within	21	0.2229	1.7294 NS
	8/14	among	2	0.0504	
		within	21	0.2077	0.2426 NS
	9/17	among	2	0.3577	
		within	21	0.2141	7.8721 **
	10/14	among	2	0.3118	
		within	21	0.1323	0.1193 NS
MINERAL	6/19	among	2	0.0348	
		within	21	0.1422	0.2450 NS
	7/16	among	2	0.2409	
		within	21	0.1077	2.2373 NS
	8/14	among	2	0.1371	
		within	21	0.0810	1.6928 NS
	9/17	among	2	0.0117	
		within	21	0.0670	0.1743 NS
	10/14	among	2	0.0056	
		within	21	0.1077	0.9491 NS
CYST COUNT					
ORGANIC	6/19	among	2	0.5732	
		within	21	0.1100	5.2107 *
	7/17	among	2	0.3264	
		within	21	0.1699	1.9212 NS
	8/14	among	2	0.9241	
		within	21	0.1041	9.8696 **
	9/17	among	2	1.4478	
		within	21	0.0933	15.4944 **
	10/14	among	2	0.4558	
		within	21	0.2775	1.6425 NS
MINERAL	6/19	among	2	0.1230	
		within	21	0.1312	0.9379 NS
	7/17	among	2	0.5044	
		within	21	0.1805	2.7940 NS
	8/14	among	2	0.2641	
		within	21	0.0250	1.1863 NS
	9/17	among	2	0.1518	
		within	21	0.1573	1.1056 NS
	10/14	among	2	0.1014	
		within	21	0.0539	1.8821 NS

Bonferroni paired t-tests for dates with significant ANOVA differences, 1990:

DATE	HORIZON	COUNT	P-VALUES		
			CON/ANT	CON/GND	ANT/GND
9/17	ORGANIC	TOTAL	*(p<.05)	**(p<.01)	NS(p>.05)
6/19	"	CYST	*(p<.05)	NS(p>.05)	NS(p>.05)
8/14	"	"	**(p<.01)	*(p<.05)	NS(p>.05)
9/17	"	"	***(p<.001)	*(p<.05)	NS(p>.05)

TABLE C-3 cont'd (ANOVA, 1991)

HORIZON	DATE	GROUPS	TOTAL COUNT		F
			DF	MS	
ORGANIC	6/10	among	2	0.1451	
		within	21	0.1605	0.9041 NS
	7/15	among	2	0.3077	
		within	21	0.0895	3.4372 NS
	8/12	among	2	0.0335	
		within	21	0.0538	0.6232 NS
	9/24	among	2	0.0754	
		within	21	0.0541	1.3927 NS
	10/20	among	2	0.0639	
		within	21	0.0914	0.6987 NS
MINERAL	6/10	among	2	0.0208	
		within	21	0.0299	0.6945 NS
	7/15	among	2	0.0673	
		within	21	0.0368	1.8284 NS
	8/12	among	2	0.0353	
		within	21	0.0402	0.8797 NS
	9/24	among	2	0.0785	
		within	21	0.0415	1.8929 NS
	10/20	among	2	0.1123	
		within	21	0.0474	2.3675 NS
CYST COUNT					
ORGANIC	6/10	among	2	0.0477	
		within	21	0.1746	0.2732 NS
	7/15	among	2	0.4963	
		within	21	0.0840	5.9105 **
	8/12	among	2	0.4891	
		within	21	0.0851	5.7465 **
	9/24	among	2	0.1089	
		within	21	0.1038	1.0499 NS
	10/20	among	2	0.4717	
		within	21	0.1436	3.2847 NS
MINERAL	6/10	among	2	0.0457	
		within	21	0.0042	3.5976 *
	7/15	among	2	0.3274	
		within	21	0.0256	8.2320 **
	8/12	among	2	0.0236	
		within	21	0.0257	0.9165 NS
	9/24	among	2	0.9652	
		within	21	0.1408	6.8536 **
	10/20	among	2	0.3802	
		within	21	0.0928	4.0985 *

Bonferroni paired t-tests for dates with significant ANOVA differences, 1991:

DATE	HORIZON	COUNT	P-VALUES		
			CON/ANT	CON/GND	ANT/GND
7/15	ORGANIC	CYST	***(p<.001)	NS(p>.05)	*(p<.05)
8/12	"	"	**(p<.01)	NS(p>.05)	**(p<.01)
6/10	MINERAL	"	NS(p>.05)	NS(p>.05)	NS(p>.05)
7/15	"	"	NS(p>.05)	**(p<.01)	*(p<.05)
9/24	"	"	*(p<.05)	NS(p>.05)	*(p<.05)
10/20	"	"	NS(p>.05)	NS(p>.05)	*(p<.05)

TABLE C-3 cont'd(ANOVA, 1992)

HORIZON	DATE	GROUPS	TOTAL COUNT		F
			DF	MS	
ORGANIC	7/20	among	2	0.5036	
		within	21	0.1535	3.2812 NS
	8/26	among	2	0.0474	
		within	21	0.0171	2.7747 NS
	9/23	among	2	0.0183	
		within	21	0.0373	0.0373 NS
	CYST COUNT				
	7/20	among	2	0.4831	
		within	21	0.1163	4.1522 *
	8/26	among	2	0.0068	
		within	21	0.0168	0.4026 NS
	9/23	among	2	0.0034	
		within	21	0.0126	0.2720 NS

Bonferroni paired t-tests for dates with significant ANOVA differences, 1992:

DATE	HORIZON	COUNT	P-VALUES		
			CON/ANT	CON/GND	ANT/GND
7/20	ORGANIC	CYST	NS(p>.05)	NS(p>.05)	*(p<.05)

APPENDIX D

In situ growth of Acanthamoeba polyphaga

TABLE D-1. 1989 regression calculations for growth of *Acanthamoeba polyphaga* in subterranean culture vessels, data log transformed. Three replicate experiments were done for both E-field and Current Density experiments at each site.

Date	Experiment	Slope	Std Error	95% Confidence Limits
6/14/89	E-Field, control	0.02333	0.00306	L1 = 0.01004 / L2 = 0.03666
	" , antenna	0.02465	0.00515	L1 = 0.00252 / L2 = 0.04679
	" , ground	0.02272	0.00592	L1 = -0.00278 / L2 = 0.04819
	Current, control	0.02008	0.00866	L1 = 0.01714 / L2 = 0.05733
	" , antenna	0.01716	0.00438	L1 = 0.00165 / L2 = 0.03602
	" , ground	0.02113	0.00510	L1 = -0.00088 / L2 = 0.04308
7/11/89	E-Field, control	0.01106	0.00188	L1 = 0.00502 / L2 = 0.01702
	" , antenna	0.00869	0.00131	L1 = 0.00459 / L2 = 0.01285
	" , ground	0.01209	0.00148	L1 = 0.00738 / L2 = 0.01681
	Current, control	0.00775	0.00150	L1 = 0.00296 / L2 = 0.01254
	" , antenna	0.00721	0.00200	L1 = 0.00083 / L2 = 0.01359
	" , ground	0.00812	0.00088	L1 = 0.00532 / L2 = 0.01092
8/18/89	E-Field, control	0.02922	0.00673	L1 = 0.00781 / L2 = 0.05063
	" , antenna	0.02886	0.00595	L1 = 0.00992 / L2 = 0.04779
	" , ground	0.02901	0.00745	L1 = 0.00529 / L2 = 0.05273
	Current, control	0.02723	0.00703	L1 = 0.00488 / L2 = 0.04959
	" , antenna	0.02912	0.00522	L1 = 0.01252 / L2 = 0.04573
	" , ground	0.02588	0.00615	L1 = 0.00633 / L2 = 0.04544

For the slope of the curve; Bonferoni T-tests of slopes revealed no significant differences::

		E-Field	Current Density
6/14/89	Control vs. Antenna	0.22049	0.30074
	Control vs. Ground	0.09155	0.10468
	Antenna vs. Ground	0.24602	0.59051
7/11/89	Control vs. Antenna	1.01051	0.21571
	Control vs. Ground	0.49665	0.12962
	Antenna vs. Ground	1.72073	0.41639
8/18/89	Control vs. Antenna	0.03952	0.21597
	Control vs. Ground	0.02042	0.13367
	Antenna vs. Ground	0.01573	0.40091

TABLE D-2. Culture cell current densities and E-field voltages measured during growth experiments (Table D-1) for June 14, 1989.

Electrodes ^t	Voc (mV)	Vcl (mV) ^u	Vr (mV)	Ecl (mV/m) ^v	Jcl (mA/m ²) ^w
Control, CD:					
1	0.98	*	0.96	*	0.002
2	0.92	*	0.95	*	0.002
3	1.09	*	1.07	*	0.003
Control, EF:					
1	0.49	0.12	*	1.06	*
2	0.60	0.12	*	1.06	*
3	0.76	0.12	*	1.06	*
Antenna, CD:					
1	51	*	51	*	0.13
2	43	*	43	*	0.14
3	56	*	56	*	0.14
Antenna, EF:					
1	15	6.4	*	56.6	*
2	17	6.4	*	56.6	*
3	20	6.4	*	56.6	*
Ground, CD:					
1	14	*	14	*	0.036
2	17	*	17	*	0.043
3	17	*	17	*	0.043
Ground, EF:					
1	7	2.00	*	17.7	*
2	10	2.00	*	17.7	*
3	10	2.00	*	17.7	*

^tCD = current density cultures; EF = E-field cultures.

^uE-field: Ecl (mV/m) = Vcl / 0.113 (length between electrodes).

^vCurrent density: Jcl (mA/m²) = Vr / R * xs. area of cl (m²), where R (ohms) = 2.5 * 10⁴ for J; 100 for E. Area of cl varied depending on submerged electrode area; for this experiment it was 1.57 * 10⁻⁴ m².

^wVcl for EF adjusted to this value, calculated: E (1m) * 0.113 (length between electrodes).

^{*}Value too low for meter to accurately record.

TABLE D-3. Culture cell current densities and E-field voltages measured during growth experiments (Table D-1) for July 11, 1989.

Electrodes ¹	Voc (mV)	Vcl (mV) ²	Vr (mV)	Ecl (mV/m) ³	Jcl (mA/m ²) ⁴
Control, CD:					
1	0.53	*	1.07	*	0.0027
2	0.58	*	0.94	*	0.0024
3	0.58	*	1.04	*	0.0026
Control, EF:					
1	1.00	0.14	*	1.2	*
2	0.86	0.14	*	1.2	*
3	1.12	0.14	*	1.2	*
Antenna, CD:					
1	32	*	32	*	0.082
2	27	*	27	*	0.069
3	33	*	33	*	0.084
Antenna, EF:					
1	16	3.76	*	33.2	*
2	16	3.76	*	33.2	*
3	16	3.76	*	33.2	*
Ground, CD:					
1	17	*	17	*	0.043
2	17	*	17	*	0.043
3	18	*	18	*	0.046
Ground, EF:					
1	20	2.13	*	18.8	*
2	12	2.13	*	18.8	*
3	11	2.13	*	18.8	*

¹CD = current density cultures; EF = E-field cultures.

²E-field: Ecl (mV/m) = Vcl / 0.113 (length between electrodes).

³Current density: Jcl (mA/m²) = Vr / R * xs. area of cl (m²), where R (ohms) = 2.5 * 10⁴ for J; 100 for E. Area of cl varied depending on submerged electrode area; for this experiment it was 1.57 * 10⁻⁴ m².

⁴Vcl for EF adjusted to this value, calculated: E (1m) * 0.113 (length between electrodes).

*Value too low for meter to accurately record.

TABLE D-4. Culture cell current densities and E-field voltages measured during growth experiments (Table D-1) for August 18, 1989.

Electrodes ¹	Voc (mV)	Vcl (mV) ¹	Vr (mV)	Ecl (mV/m) ²	Jcl (mA/m ²) ³
Control, CD:					
1	1.43	*	1.44	*	0.0037
2	1.3	*	1.29	*	0.0033
3	1.57	*	1.64	*	0.0042
Control, EF:					
1	1.4	0.2	*	1.77	*
2	1.58	0.2	*	1.77	*
3	1.5	0.2	*	1.77	*
Antenna, CD:					
1	53	*	53	*	0.13
2	44	*	44	*	0.11
3	54	*	54	*	0.14
Antenna, EF:					
1	86	6.18	*	54.7	*
2	94	6.18	*	54.7	*
3	71	6.18	*	54.7	*
Ground, CD:					
1	22	*	22	*	0.056
2	21	*	21	*	0.054
3	24	*	24	*	0.061
Ground, EF:					
1	27	2.72	*	24.1	*
2	36	2.72	*	24.1	*
3	14	2.72	*	24.1	*

¹CD = current density cultures; EF = E-field cultures.

²E-field: Ecl (mV/m) = Vcl / 0.113 (length between electrodes).

³Current density: Jcl (mA/m²) = Vr / R * xs. area of cl (m²), where R (ohms) = 2.5 * 10⁶ for J; 100 for E. Area of cl varied depending on submerged electrode area; for this experiment it was 1.57 * 10⁻⁴ m².

'Vcl for EF adjusted to this value, calculated: E (1m) * 0.113 (length between electrodes).

*Value too low for meter to accurately record.

TABLE D-5. 1990 regression calculations for growth of *Acanthamoeba polyphaga* in subterranean culture vessels, data log transformed. Three replicate experiments were done for both E-field and Current Density experiments at each site.

Date	Experiment	Slope	Std Error	95% Confidence Limits
6/11/90	E-Field, control	0.03139	0.00297	L1= 0.00634/L2= 0.06912
	" , antenna	0.02797	0.00166	L1= 0.00694/L2= 0.04901
	" , ground	0.03114	0.00610	L1=-0.04642/L2= 0.10869
	Current, control	0.02895	0.00654	L1=-0.05409/L2= 0.11199
	" , antenna	0.02641	0.00514	L1=-0.03884/L2= 0.09165
	" , ground	0.03155	0.00168	L1= 0.01008/L2= 0.05301
7/10/90	E-Field, control	0.01542	0.00426	L1=-0.00292/L2= 0.03371
	" , antenna	0.01355	0.00216	L1= 0.00423/L2= 0.02287
	" , ground	0.01338	0.00316	L1= 0.00021/L2= 0.02696
	Current, control	0.01755	0.00171	L1= 0.01017/L2= 0.02492
	" , antenna	0.01213	0.00239	L1= 0.00181/L2= 0.02244
	" , ground	0.01428	0.00412	L1=-0.00347/L2= 0.03204
7/30/90	E-Field, control	0.01811	0.00152	L1= 0.01568/L2= 0.02464
	" , antenna	0.01927	0.00168	L1= 0.01203/L2= 0.02651
	" , ground	0.01860	0.00138	L1= 0.01262/L2= 0.02458
	Current, control	0.01984	0.00098	L1= 0.01559/L2= 0.02408
	" , antenna	0.01841	0.00058	L1= 0.01588/L2= 0.02093
	" , ground	0.01838	0.00181	L1= 0.01059/L2= 0.02616

For the slope of the curve, Bonferoni T-tests of slopes revealed no significant differences

		E-Field	Current Density
6/11/90	Control vs. Antenna	1.00467	0.30060
	Control vs. Ground	0.03734	0.38500
	Antenna vs. Ground	0.50005	0.48100
7/10/90	Control vs. Antenna	0.42818	1.83853
	Control vs. Ground	0.38600	0.73079
7/30/90	Antenna vs. Ground	0.45065	0.45075
	Control vs. Antenna	0.51254	1.24954
	Control vs. Ground	0.24034	0.70934
	Antenna vs. Ground	0.30576	0.01495

TABLE D-6. Culture cell current densities and E-field voltages measured during growth experiments (Table D-5) for June 11, 1990.

Electrodes ¹	Voc (mV)	Vcl (mV) ²	Vr (mV)	Ecl (mV/m) ³	Jcl (mA/m ²) ⁴
Control, CD:					
1	1.64	*	1.65	*	0.004
2	1.44	*	1.44	*	0.004
3	1.77	*	1.80	*	0.005
Control, EF:					
1	2.43	0.21	*	1.86	*
2	1.94	0.21	*	1.86	*
3	2.36	0.21	*	1.86	*
Antenna, CD:					
1	52	*	51	*	0.13
2	46	*	44	*	0.11
3	56	*	55	*	0.14
Antenna, EF:					
1	59	6.5	*	57.2	*
2	95	6.6	*	59.5	*
3	59	6.6	*	58.6	*
Ground, CD:					
1	19	*	20	*	0.052
2	23	*	13	*	0.033
3	23	*	20	*	0.052
Ground, EF:					
1	26	3.40	*	30.1	*
2	28	3.50	*	31.0	*
3	14	3.60	*	31.9	*

¹CD = current density cultures; EF = E-field cultures.

²E-field: Ecl (mV/m) = Vcl / 0.113 (length between electrodes).

³Current density: Jcl (mA/m²) = Vr / R * xs. area of cl (m²), where R (ohms) = 2.5×10^4 for J; 100 for E. Area of cl varied depending on submerged electrode area; for this experiment it was 1.57×10^{-4} m².

⁴Vcl for EF adjusted to this value, calculated: E (1m) * 0.113 (length between electrodes).

*Value too low for meter to accurately record.

TABLE D-7. Culture cell current densities and E-field voltages measured during growth experiments (Table D-5) for July 10, 1990.

Electrodes ¹	Voc (mV)	Vcl (mV) ²	Vr (mV)	Ecl (mV/m) ³	Jcl (mA/m ²) ⁴
Control, CD:					
1	1.4	*	1.35	*	0.003
2	1.2	*	1.26	*	0.003
3	1.4	*	1.43	*	0.004
Control, EF:					
1	1.9	0.16	*	1.4	*
2	1.6	0.16	*	1.4	*
3	1.9	0.16	*	1.4	*
Antenna, CD:					
1	49	*	48	*	0.12
2	45	*	44	*	0.11
3	54	*	54	*	0.14
Antenna, EF:					
1	59	6.4	*	56.6	*
2	92	6.7	*	59.3	*
3	60	6.5	*	57.5	*
Ground, CD:					
1	19	*	21	*	0.055
2	19	*	20	*	0.051
3	22	*	24	*	0.061
Ground, EF:					
1	22	3.1	*	27.5	*
2	25	2.7	*	24.1	*
3	21	3.3	*	29.6	*

¹CD = current density cultures; EF = E-field cultures.

²E-field: Ecl (mV/m) = Vcl / 0.113 (length between electrodes).

³Current density: Jcl (mA/m²) = Vr / R * xs. area of cl (m²), where R (ohms) = 2.5 * 10⁶ for J; 100 for E. Area of cl varied depending on submerged electrode area; for this experiment it was 1.57 * 10⁻⁴ m².

⁴Vcl for EF adjusted to this value, calculated: E (1m) * 0.113 (length between electrodes).

*Value too low for meter to accurately record.

TABLE D-8. Culture cell current densities and E-field voltages measured during growth experiments (Table D-5) for July 30, 1990.

Electrodes ¹	Voc (mV)	Vcl (mV) ²	Vr (mV)	Ecl (mV/m) ³	Jcl (mA/m ²) ⁴
Control, CD:					
1	2	*	2.1	*	0.005
2	2	*	1.8	*	0.005
3	2	*	2.2	*	0.006
Control, EF:					
1	3	0.26	*	2.3	*
2	2	0.26	*	2.3	*
3	3	2.8	*	25	*
Antenna, CD:					
1	52	*	51	*	0.13
2	46	*	46	*	0.12
3	53	*	53	*	0.14
Antenna, EF:					
1	61	5.7	*	50.2	*
2	89	6.3	*	55.3	*
3	61	6.4	*	56.5	*
Ground, CD:					
1	19	*	24	*	0.06
2	24	*	23	*	0.06
3	25	*	22	*	0.06
Ground, EF:					
1	24	3.1	*	27.3	*
2	25	2.9	*	25.8	*
3	17	3.1	*	27.4	*

¹CD = current density cultures; EF = E-field cultures.

²E-field: Ecl (mV/m) = Vcl / 0.113 (length between electrodes).

³Current density: Jcl (mA/m²) = Vr / R * xs. area of cl (m²), where R (ohms) = 2.5 * 10⁴ for J; 100 for E. Area of cl varied depending on submerged electrode area; for this experiment it was 1.57 * 10⁻⁴ m².

⁴Vcl for EF adjusted to this value, calculated: E (1m) * 0.113 (length between electrodes).

*Value too low for meter to accurately record.

TABLE D-9. 1991 regression calculations for growth of *Acanthamoeba polyphaga* in subterranean culture vessels, data log transformed. Three replicate experiments were done for both E-field and Current Density experiments at each site.

Date	Experiment	Slope	Std Error	95% Confidence Limits
6/24 to 6/28/91	E-Field, control	0.01607	0.00062	L1= 0.01337/L2= 0.01877
	" , antenna	0.01542	0.00129	L1= 0.00985/L2= 0.02099
	" , ground	0.01558	0.00277	L1= 0.03631/L2= 0.02753
	Current, control	0.01321	0.00215	L1= 0.00394/L2= 0.02249
	" , antenna	0.01843	0.00086	L1= 0.01469/L2= 0.02216
	" , ground	0.01491	0.00228	L1= 0.00505/L2= 0.02476
7/29 to 8/2/91	E-Field, control	0.01868	0.00141	L1= 0.00085/L2= 0.03651
	" , antenna	0.01569	0.0004	L1= 0.01060/L2= 0.02078
	" , ground	0.02035	0.00204	L1= -0.00563/L2= 0.04633
	Current, control	0.02069	0.00032	L1= 0.01662/L2= 0.02477
	" , antenna	0.01444	0.00561	L1= -0.05687/L2= 0.08576
	" , ground	0.01868	0.00285	L1= -0.01748/L2= 0.05485
8/19 to 8/23/91	E-Field, control	0.01063	0.00031	L1= 0.00670/L2= 0.01456
	" , antenna	0.01118	0.00061	L1= 0.00335/L2= 0.01882
	" , ground	0.01036	0.00031	L1= 0.00642/L2= 0.01428
	Current, control	0.01074	0.00335	L1= -0.03183/L2= 0.05332
	" , antenna	0.01001	0.00115	L1= 0.00046/L2= 0.02464
	" , ground	0.01074	0.00022	L1= 0.00791/L2= 0.01358

For the slope of the curve, Bonferoni T-tests of slopes revealed no significant differences

		E-Field	Current Density
6/24/91	Control vs. Antenna	0.45415	2.25426
	Control vs. Ground	0.17362	0.54247
	Antenna vs. Ground	0.05236	1.44452
7/29/	Control vs. Antenna	2.0401	1.11228
	Control vs. Ground	0.67343	0.70086
	Antenna vs. Ground	2.241	1.19839
8/19/91	Control vs. Antenna	0.80379	0.20611
	Control vs. Ground	0.61587	0.00001
	Antenna vs. Ground	1.19839	0.62348

TABLE D-10. Culture cell current densities and E-field voltages measured during growth experiments (Table D-9) for June 26, 1991.

				(mV/m) ¹	(mA/m ²) ²
Control,CD:					
1	2.37	*	2.32	*	0.006
2	2.24	*	2.29	*	0.006
3	2.76	*	2.71	*	0.008
Control,EF:					
1	3.69	0.32	*	2.83	*
2	2.87	0.32	*	2.83	*
3	3.68	0.32	*	2.83	*
Antenna,CD:					
1	54	*	36	*	0.09
2	46	*	46	*	0.12
3	53	*	53	*	0.14
Antenna,EF:					
1	64	6.3	*	53.8	*
2	89	6.2	*	54.9	*
3	62	6.2	*	54.9	*
Ground,CD:					
1	22	*	21	*	0.05
2	22	*	21	*	0.05
3	24	*	22	*	0.06
Ground,EF:					
1	27	3.4	*	28.8	*
2	24	2.8	*	24.8	*
3	16	3.7	*	32.7	*

¹CD = current density cultures; EF = E-field cultures.

²E-field: $E_{cl} \text{ (mV/m)} = V_{cl} / 0.113$ (length between electrodes).

³Current density: $J_{cl} \text{ (mA/m}^2\text{)} = V_r / R * \text{xs. area of cl (m}^2\text{)}$, where $R \text{ (ohms)} = 2.5 * 10^3$ for J; 100 for E. Area of cl varied depending on submerged electrode area; for this experiment it was $1.57 * 10^{-4} \text{ m}^2$.

⁴ V_{cl} for EF adjusted to this value, calculated: $E \text{ (Vm)} * 0.113$ (length between electrodes).

*Value too low for meter to accurately record.

TABLE D-11. Culture cell current densities and E-field voltages measured during growth experiments (Table D-9) for July 29, 1991.

Electrodes:	Voc (mV)	Vcl (mV) ^a	Vr (mV)	Ecl (mV/m) ^b	Jcl (mA/m ²) ^c
Control, CD:					
1	1	*	1	*	0.003
2	0.9	*	0.9	*	0.002
3	1.1	*	1	*	0.003
Control, EF:					
1	1.4	0.14	*	1.24	*
2	1.2	0.14	*	1.24	*
3	1.4	0.14	*	1.24	*
Antenna, CD:					
1	51	*	50	*	0.13
2	43	*	43	*	0.11
3	55	*	55	*	0.14
Antenna, EF:					
1	61	6.1	*	54	*
2	92	6.3	*	56	*
3	62	6.3	*	56	*
Ground, CD:					
1	22	*	21	*	0.05
2	22	*	22	*	0.06
3	22	*	22	*	0.06
Ground, EF:					
1	23	3.1	*	27	*
2	26	2.9	*	26	*
3	17	2.6	*	23	*

^aCD = current density cultures; EF = E-field cultures.

^bE-field: Ecl (mV/m) = Vcl / 0.113 (length between electrodes).

^cCurrent density: Jcl (mA/m²) = Vr / R * xs. area of cl (m²), where R (ohms) = 2.5 * 10⁶ for J; 100 for E. Area of cl varied depending on submerged electrode area; for this experiment it was 1.57 * 10⁻⁴ m².

^aVcl for EF adjusted to this value, calculated: E (1m) * 0.113 (length between electrodes).

^aValue too low for meter to accurately record.

TABLE D-12. Culture cell current densities and E-field voltages measured during growth experiments (Table D-9) for August 21, 1991.

Electrodes ¹	Voc (mV)	Vcl (mV) ¹	Vr (mV)	Ecl (mV/m) ²	Jcl (mA/m ²) ³
Control, CD:					
1	1.3	*	1.2	*	0.003
2	1.1	*	1.2	*	0.003
3	1.3	*	1.3	*	0.003
Control, EF:					
1	2	0.16	*	1.42	*
2	1.5	0.16	*	1.42	*
3	1.9	0.16	*	1.42	*
Antenna, CD:					
1	52	*	51	*	0.13
2	45	*	45	*	0.11
3	53	*	54	*	0.14
Antenna, EF:					
1	64	6.5	*	58	*
2	91	6.1	*	54	*
3	60	6.4	*	57	*
Ground, CD:					
1	22	*	21	*	0.05
2	24	*	23	*	0.06
3	24	*	23	*	0.06
Ground, EF:					
1	26	3.3	*	29	*
2	29	3.6	*	32	*
3	17	3.4	*	30	*

¹CD = current density cultures; EF = E-field cultures.

²E-field: Ecl (mV/m) = Vcl / 0.113 (length between electrodes).

³Current density: Jcl (mA/m²) = Vr / R * xs. area of cl (m²), where R (ohms) = 2.5 * 10⁴ for J; 100 for E. Area of cl varied depending on submerged electrode area; for this experiment it was 1.57 * 10⁻⁴ m².

'Vcl for EF adjusted to this value, calculated: E (1m) * 0.113 (length between electrodes).

*Value too low for meter to accurately record.

The following procedures were used for testing growth of amoebae in electric fields, provided by the IIT Research Institute

MATCHED E-FIELD PROTOCOL

- 1) Measure maximum E-field in soil using 1 meter probe- E.
 - 2) Multiply E-field value by 0.15 to determine the minimum required drive voltage, V_{DR} (min).
- $$V_{DR} \text{ (min)} = E \times 0.15 \text{ (volts)}$$
- 3) Locate collector electrodes in line with the maximum E-field in the earth, and spaced far enough apart to generate a voltage across a 2000 ohm resistor which is greater than or equal to V_{DR} (min). See Figure 1.
 - 4) Measure and record electrode spacing and the open circuit (no lead) electrode voltage, V_{oc} .
 - 5) Connect the test cell and monitoring box to the electrodes. Refer to Figure 2. While monitoring the voltage across the test cell only, V_{CL} , adjust the variable resistor so that the cell voltage is equal to the value given by the following formula:

$$V_{CL} = E \times 0.113 \text{ (volts)}$$

- 6) With the cell voltage set, measure and record the voltage across the 100 ohm series resistor, V_R . This allows calculation of the cell current and current density.
- 7) Measure and record the electrode voltage with the test cell and monitoring box connected and adjusted as per Step 5, V_{DR} .

MATCHED CURRENT DENSITY PROTOCOL

- 1) Measure maximum E-field in soil using 1 meter probe -E.
- 2) Locate collector electrodes in line with maximum E-field with a separation of 1 meter.
- 3) Measure exact electrode spacing and open circuit (no load) electrode voltage, V_{oc} . Measured voltage should be within a few percent of that measured in Step 1. If not, correct electrode spacing as appropriate.
- 4) Connect current-limiting test chamber (see Figure 3) to electrodes. Place the current limit select switch to the 250 mesohm position 2.5M.

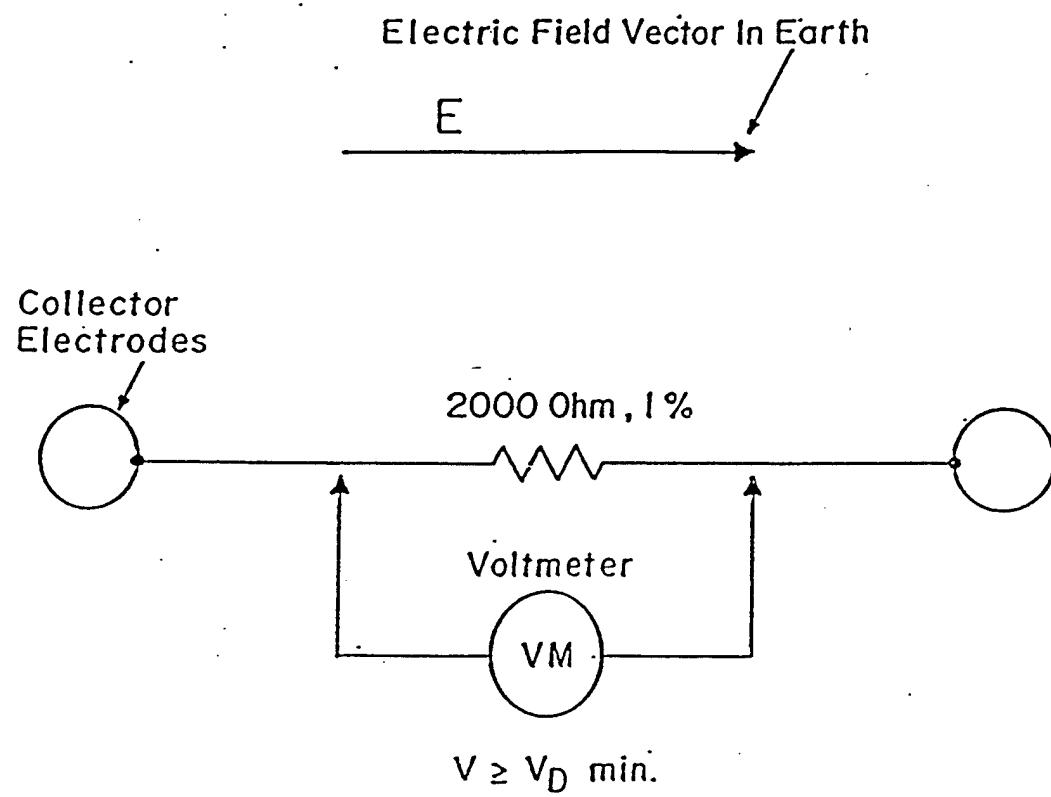
- 5) Measure and record the voltages across the test cell, V_{CL} , the resistor, V_R , and the electrodes V_{DR} , using the test point jacks. Refer to Figure 3 for test point numbering.

The voltages across the resistor and across the electrodes should be close in value to V_{OC} from Step 3.

$$V_R \approx V_{DR} \approx V_{OC}$$

The voltage across the test cell will be much lower, and can be estimated as:

$$V_{CL} = 0.6 \times 10^{-3} \times V_c \text{ (volts)}$$



PLANE VIEW

FIGURE D-1. Determination of drive voltage, step 3 in the matched E-field protocol.

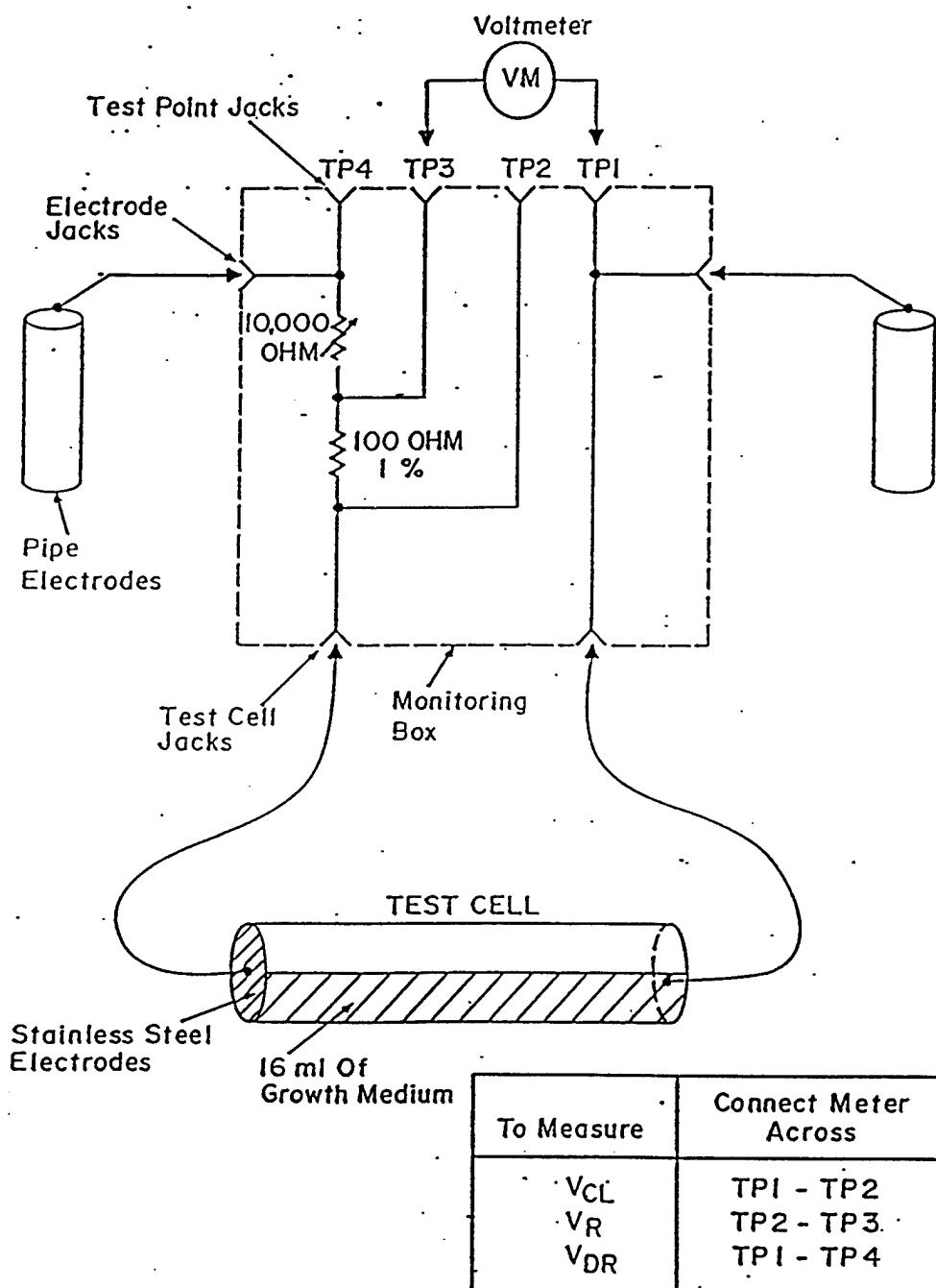


FIGURE D-2. Test cell hookup for matched E-field protocol, step 5.

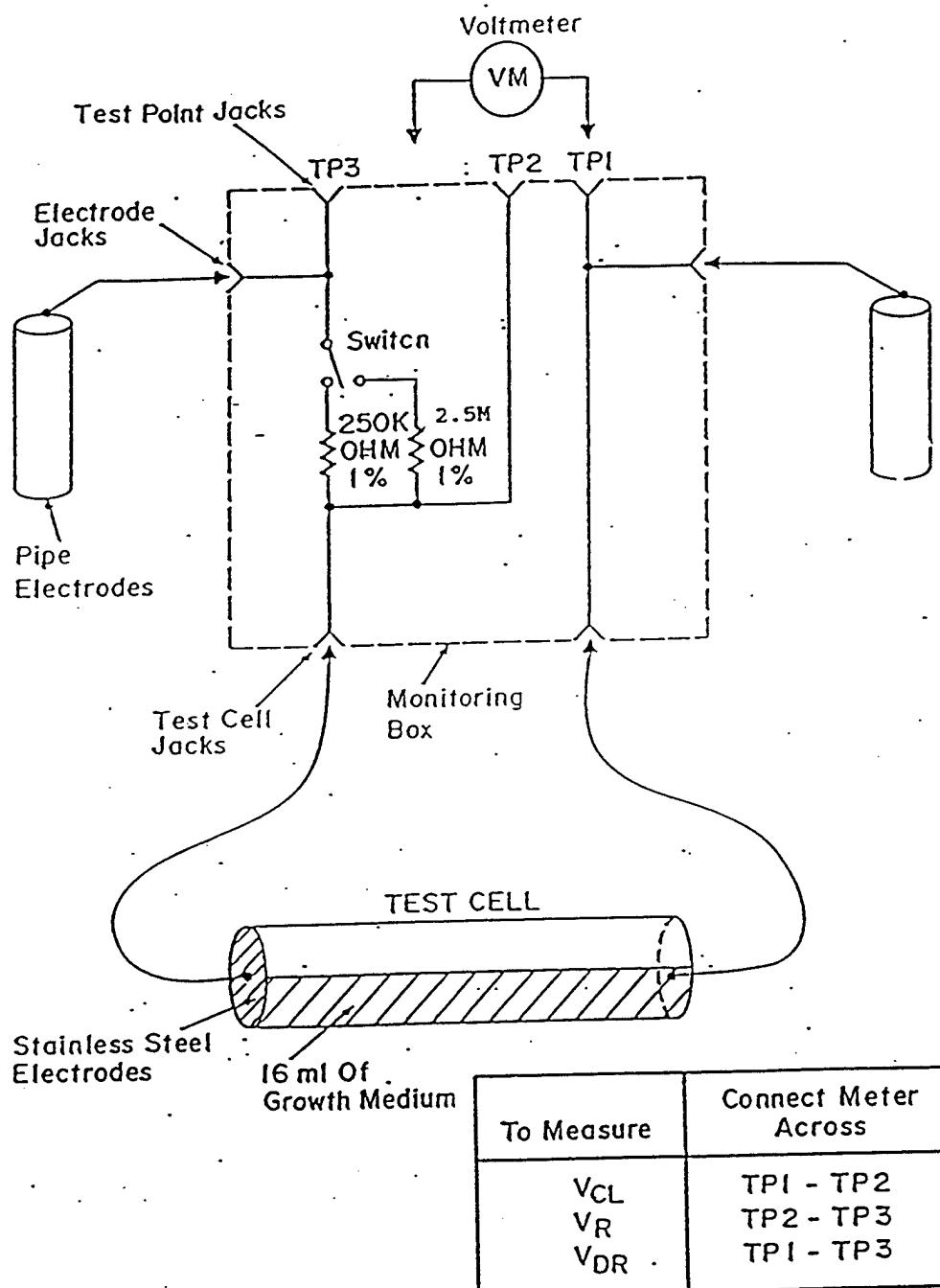


FIGURE D-3. Test cell hookup for matched current density protocol, step 4.

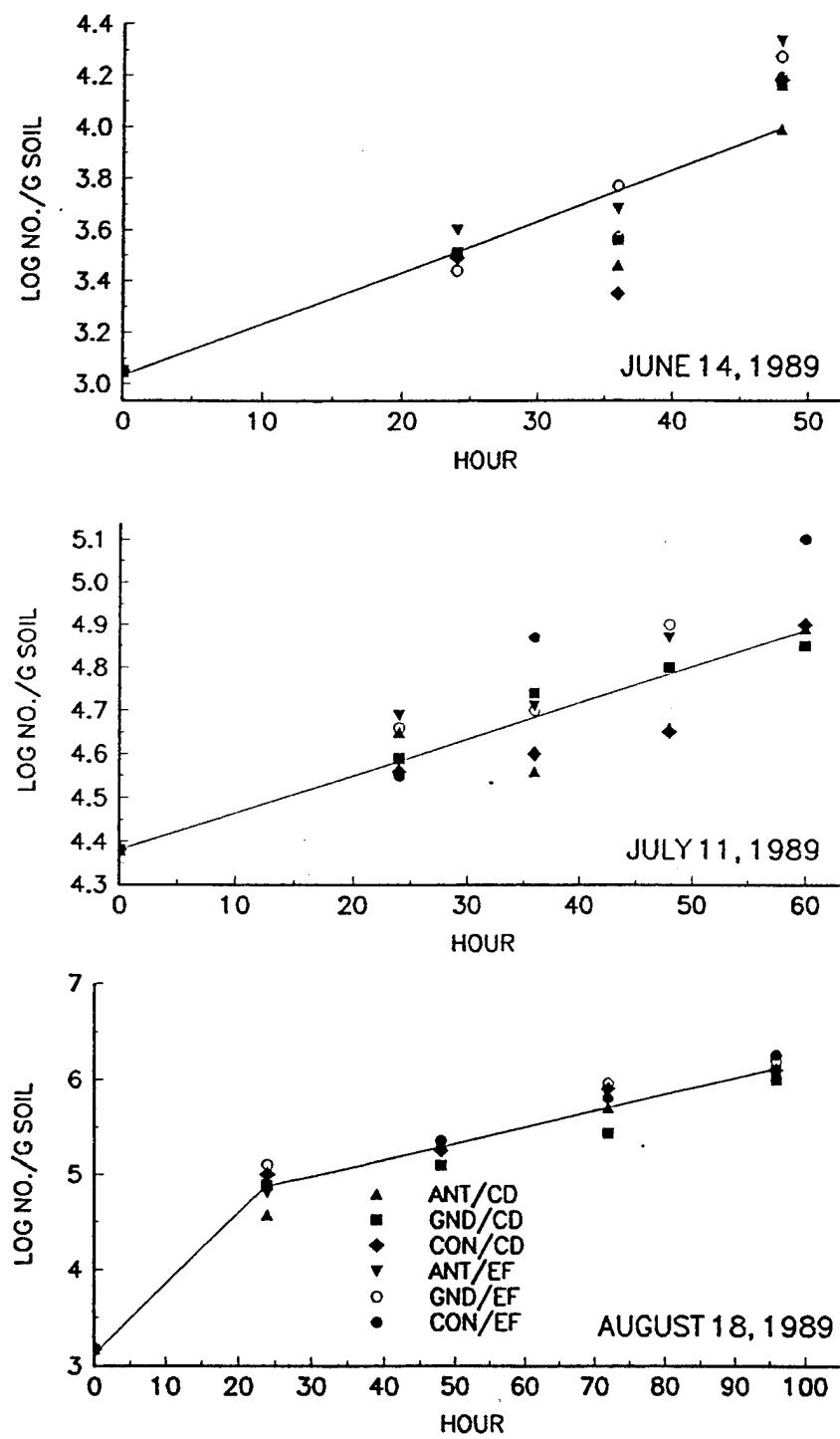


FIGURE D-4. Growth in buried culture vessels. Points represent means, $n = 3$, from cultures at ANT, GND and CON sites, subject to current density (CD) and E-field voltages (EF) found in soil at the sites. Data for 1989.

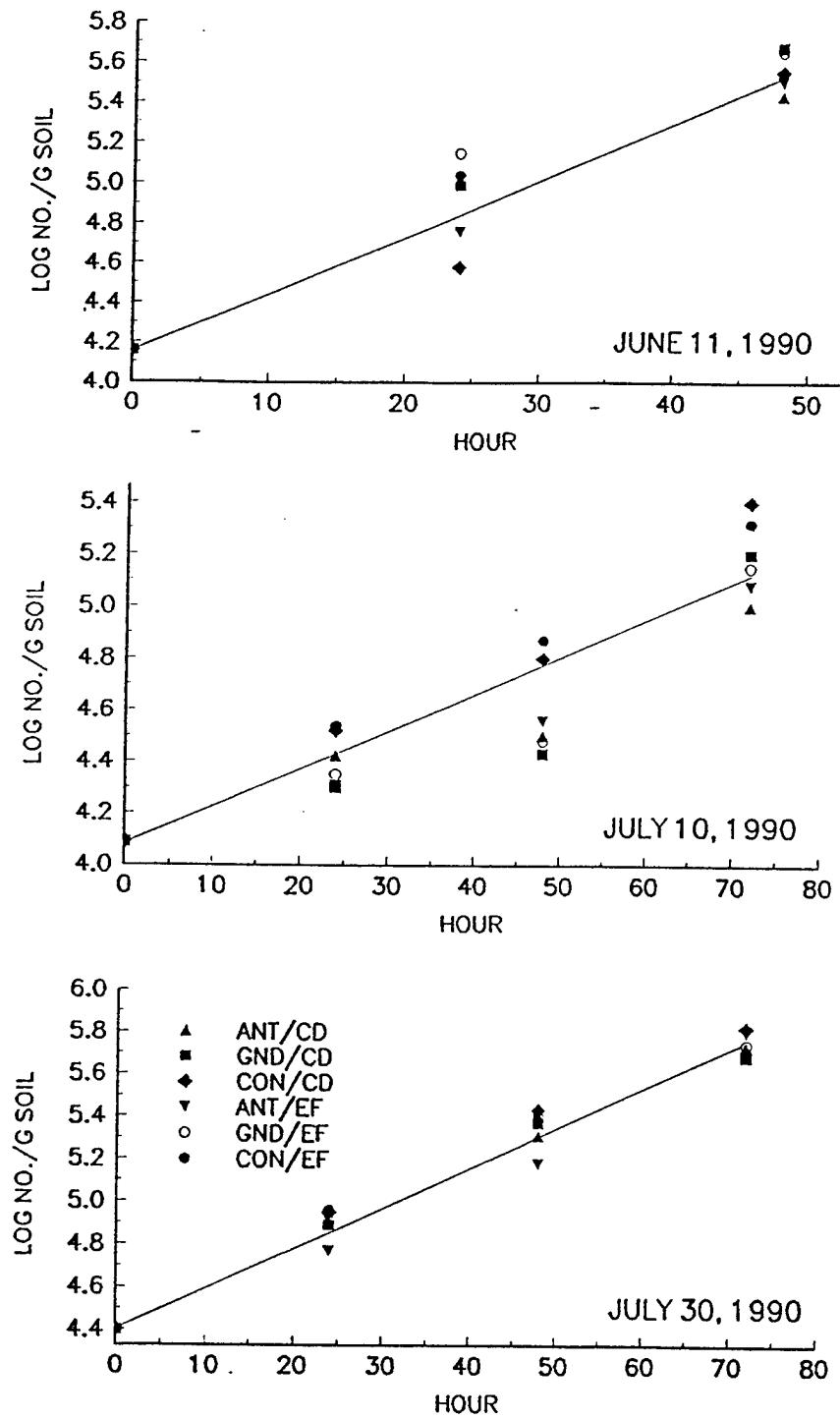


FIGURE D-5. Growth in buried culture vessels. See Fig. D-4, data for 1990.

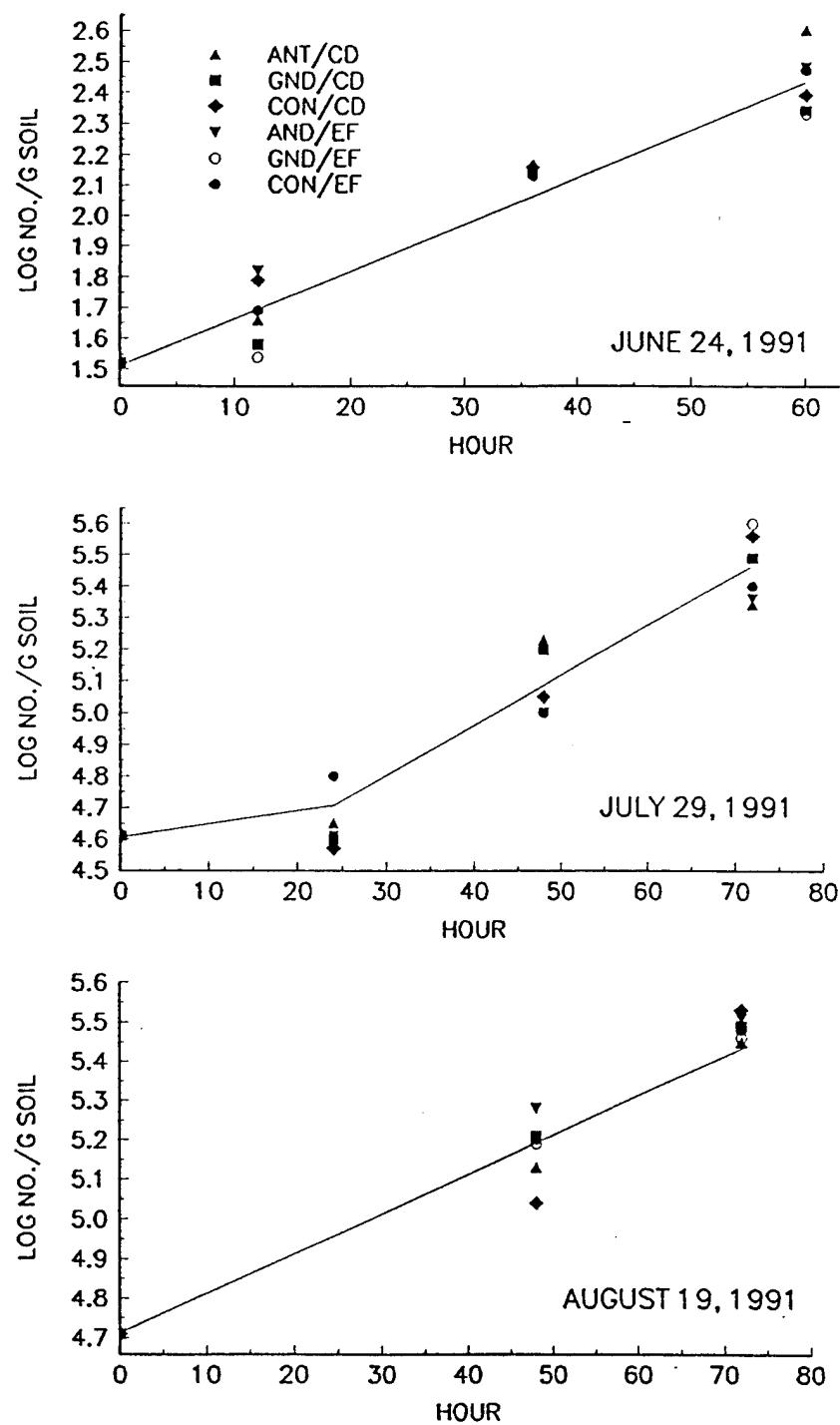


FIGURE D-6. Growth in buried culture vessels. See Fig. D-4, data for 1991.

APPENDIX E
Allozyme Data

TABLE E-1. Allozyme gel data, control site, 1993. Clone numbers given on horizontal axis, allozyme loci given on vertical axis. Allozyme symbols given in Table 9 of report.

LOCI	6	7	9	10	14	19	21	23	25	28	32	38	39	40	41
AE1	1/3	1/2	3	4/5	1/4	3	1/2	1/2	1	1/2	1/2	4/5	1/2	1/2	3
2	1/2	1/2	1/2	1/3	1/2	1/2	1/3	1/3	1/2	1/2	1/3	1	1/3	1/3	1/3
3	1/4	1/5	1/2	1	1/2	1/4	1/3	1/3	1/2	1/2	1/2	1/2	1/3	1/2	1/2
4	1/2	1/3	1/2	1/2	2/4	1/2	4/5	1/2	3/4	1/2	1/2	1/3	1/2	1/2	1/2
PE1	3/4	1/2	3	3/4	3/4	2/3	1	1/3	1	3	3/4	4/5	1	1	3
2	1/2	1/2	1/3	1/2	1/3	1/2	1/2	1/2	1/2	1/2	1/3	1	1/2	1/2	1/2
3	1	1/2	1/2	1/2	1/3	1/2	1/3	1/2	1	1/2	1/3	1/2	1/2	1/2	1/4
4	1	1/2	1/2	1/2	2	1/2	1/2	1/2	1/2	1/2	2	1/3	1/2	1/2	1/3
5	3	1/2	2	1/2	1	2	1/2	1/2	2	2	1	1/2	1/2	1	1/2
BE1	1/3	1/3	1/3	4/5	1/2	3/4	1	1	1/2	4	1/2	4/5	1	1	3/4
2	1/2	1/2	1/3	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/3
3	1	1/3	1/2	1/2	1	1/2	1	1	1/2	3	2	3	1/2	1	3/4
4	1/2	1/2	2	2	1	2	1	1	2	2	1	2	1	1	1/2
5	1	2	1/2	1/2	1/2	2	1	1/2	1/2	1/2	1/2	1/2	1/2	1	1/2
SOD1	1/3	2	1/2	1	1/2	1	1/2	1/2	1	1/3	1/3	1	1/2	1/2	1
2	3	1/3	3/4	1	3/4	1/3	3	3/4	4	3/4	3	1	3	3/4	3/4
3	1/2	1/2	1/4	1/2	1/3	1/2	1/2	1/2	1	1/2	1/2	1/2	1/3	1/2	1/2
ACP1	1/2	1/2	1/4	1/2	1/2	1/2	1/2	1/2	2/3	1/4	1/3	1/2	1/2	1/2	1/4
2	2	1/2	1/4	2	2	1/2	1/2	2	3	4	3	1/2	1	2	1/3
3	2	1/2	1/2	1/3	2	1	1/4	1/4	1/3	1/4	2	1/4	1	1/4	1/2
GDH1	1	1	1/2	1/4	1	1/2	1/4	1/3	1/4	2	1/2	1	1/4	1/4	2
LTD1	4	3	1	1	2	1	1	1/3	3	1	1/2	1	1	1/2	1
2	1/2	1/4	1	1/2	1	1	1/2	1/2	1/2	1	2	1/2	1/3	1/3	1
LDH1	2	1	1/2	1	2	1/2	1	1/2	1	1	2	1/2	1/2	1/2	1
2	1/2	2	1	1/3	1	1/3	1/2	1/4	1	1	1	1/3	1/2	1/3	1
PGM1	1/2	1	1	1	1	1/2	1	1/2	1	1	1	1	1/2	1/2	1
2	1	1/2	1	1/2	1	1	1	1/2	1	1	1	1	1	1	1
3	1/2	3	1/3	1	2	1/2	1/2	1/2	1/3	1	2	2	1/2	1	1

TABLE E-1 cont'd (control site).

LOCI	55	56	58	66	72	77	88	89	81	82	83	84	85	86	87
AE1	2	2	1/2	1/2	1/2	1/2	1/3	4/5	1/2	1	1/4	1	1/4	1/3	1/3
2	0	0	1/2	1/2	4/5	2	2	1/4	1/3	1/2	3/4	1/3	3/4	1/2	1/2
3	1/3	1/3	1/2	1/2	2	3	1/3	1/4	1/2	1/2	4	1/3	4	1/3	1/2
4	1/2	1/2	1/3	4/5	1/3	1/2	1/2	1/2	1/2	3/4	1/2	1/2	1/2	1/2	1/2
PE1	1	1	2/4	1	1	1	1/3	4	1	1/3	4/8	1/3	4/8	1/3	1
2	0	0	2/3	1/2	1	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2
3	2	2	1/2	1/2	4	1/2	1/3	1/3	1/2	1/3	1/2	1/2	1/2	1/3	1/2
4	4	4	1/3	1/2	1/3	1/2	1/2	1/2	1/2	1/3	1/2	1/2	1/2	1/3	1/2
5	2	2	2	1	1/2	1	1/2	1/2	1	1/2	1/2	1/2	1/2	1/2	1/2
BE1	1	1/2	2	1	1	1/3	1	4/5	1	1/2	1/2	1	3	1/2	1
2	1/2	1/2	1/2	1/3	1/2	1/2	1/2	1/2	1/3	1/2	1/3	1/3	1/3	1/2	1/3
3	2	1/3	2	1/3	1	1	2	2	2	1	1/3	1	1/3	1	1/2
4	1/2	1/4	1/2	1/2	1	1	1/2	2	1/2	1	1	1/2	1	1	1/2
5	1/2	1	1/2	1	1/2	1/2	1/2	1/2	1	1/2	1/2	1/2	1/2	1/2	1
SOD1	3	3	1/3	1/2	1/2	1	1/2	1	1/2	1	1/4	3/4	1/4	1/2	1/2
2	1	1	3	3	3	4	1	2	3	1/2	1/2	3	1/2	1/2	3
3	1	1	1/2	1/2	1/2	1/2	1/2	1/2	1/3	1/2	1/2	1/2	1/2	1/2	1/3
ACP1	1	1	1/3	1/2	1	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/3	1/2
2	1	1	3	1/3	2	1/2	1/2	1	1/2	1/2	1	1/2	1	1/2	1/2
3	2	2	2	1/4	1	3	3	1/3	1/3	1/4	2	1/3	1/3	3	1/4
GDH1	2	2	1/2	1/2	2	1	1	1/4	1	2	1	1/3	1	1	1/4
LTD1	3	3	1/2	1/5	1/2	1/2	1/2	1/2	1	1/2	1/4	1	1/4	1/2	1
2	1/2	1/2	1/2	1	1	0	2/3	1/2	1	1/3	1/2	2	1	2/3	1
LDH1	1	1/2	2	1	1/2	1/2	1	1	1	1/2	1/2	1/3	1/2	1	1/2
2	1/3	1	1	1/4	1/2	1	1/2	1	1	1/2	1	1/2	1	1/2	1/2
PGM1	1	1	1/2	1	1	2	1/3	2	1	1/2	1/2	1	1/3	1/2	1
2	1/3	1/3	1	1	1	1	1	1	1	1	1	1	1	1	1/2
3	1/3	3	2	1/4	1/2	2	2	1	1	2	3/4	3	2/3	2	1/2

TABLE E-2. Allozyme gel data, antenna site, 1993. See Table E-1 for details.

LOCI	94	2	3	4	11	12	13	26	31	36	50	51	52	54	59
AE1	1/2	1/2	1/3	1/3	1/2	1/2	1/2	1/2	1/2	1/2	1/5	1/5	2/3	2/3	1/3
2	1/3	1/3	1/2	2/3	1/2	1/3	1/2	1/2	1/3	1/2	1/2	1/2	1/2	1/2	1
3	4/5	1	1/2	1/2	1	1/3	1	1/2	1/2	1/2	1/3	1/4	1/2	1/2	1
4	1/2	1/2	1/2	1/2	1/2	1/2	1/2	4/5	1/2	1/2	1/2	1/2	2/4	2/4	1/2
PE1	1/3	1	3/4	1	1/3	1/3	1	1/2	1/3	3	1	1	1/3	1/3	1
2	1/3	1	1/3	1	1/3	1/3	1/2	1/3	1/3	1/2	1/3	1/3	1/2	1/3	1/3
3	1	1/2	1/2	1/3	1/2	1/2	1	1/2	1/3	1/2	1/3	1/4	1/3	1	1/3
4	1/2	1/2	1/2	2	1/2	1/2	1/2	1/2	1/3	1/2	3/4	3/4	1/2	1/2	2
5	1/2	1/2	1/2	1/2	1	1	1/2	1/2	1/2	1/2	2	1/2	2	2	1
BE1	1/2	1/2	1	1	1/2	1/2	1/2	1	1/2	3/4	1	1	2	2	1
2	1/2	1	1/2	2	1	1/3	1/3	1/3	1/2	1/2	1	1	1/2	1	1/2
3	1	1	1	3	1/2	1	1/2	1/3	1	3	3/4	3/4	3	1/2	1
4	1/2	1/2	1/3	1	1	1	2	1/2	1/2	2	1/3	1/3	2	1	1
5	1/2	1	1	1/2	1	1	1/2	1	1/2	1/2	1/2	1/2	1/2	2	1
SOD1	3	1/3	1/3	1	3	3	1	3/4	3/4	1/3	3	3	3	1	3
2	3	3/4	3	3	3	3	1/2	4	3	3/4	1	1	1	3	3
3	3/4	1/3	1/3	1/2	1/2	1/2	1/2	1	1/2	1/2	1	1	3	1	1/3
ACP1	1/2	1/2	1/2	3	1/2	1/2	1/2	1/4	1/2	1/4	2/3	1	1	3/4	1/2
2	2	2	2	2	1	1	1	3	1/2	4	1/2	1	1	3/4	2
3	1/3	2	2	2	1/3	1/3	1/2	2	1/3	1/4	1/2	1/2	2	1/4	1/3
GDH1	1/3	1	1/2	0	1/2	1/2	1	3	2	1	1/2	1/4	3	1/2	1/5
LTD1	1	1/3	1/3	1/2	1	1	3	1/2	1	3/4	1/2	1	2	2	1
2	1	1/2	1/2	1/4	2	1	1/2	2	1	1	1/2	1	1/2	1/2	1/2
LDH1	1	1	1	1/2	1/2	1	1	1	1	1/2	1/2	1	1/2	1	1
2	1/2	1/2	1/2	1/4	1/2	1	1	1	1/2	1	1	1/4	1/3	1/4	3/4
PGM1	3	1/2	1/2	1/2	1/2	1/3	1	1	2	1	1	1/2	1	1/2	1
2	1	1	1	1	1	1	1	1	1	1	1/2	1/2	1/2	1	1
3	3	1/2	1/2	1/2	1/2	1/2	4	2/3	1/2	3	1	1	1	2	1/2

TABLE E-2 cont'd (antenna site).

LOCI	60	61	70	71	75	97	98	99	100	101	104	118	119	120	121
AE1	1/2	1/3	1	1/2	1/2	1/5	1/2	4/5	1/5	1/2	1/2	3/4	1/2	1/2	1/2
2	3	1/3	1/3	1/2	1/2	1/2	1/2	1	2/3	1/2	1/2	1/2	1/2	1/2	1/2
3	2/3	1/4	1/2	1/3	1/2	1/2	1/3	1/2	4	4/5	1/3	1	2/3	1/5	2/3
4	1/2	1/2	3/4	2/4	1/2	3/4	1/2	2/4	1/2	1/2	1/2	2/4	1/2	1/2	1/2
PE1	3/4	5/6	1/3	2/3	1	3/4	2	3/4	7/8	1/3	1/3	2/3	2	3/4	4
2	1	1/2	1/3	1/3	1/3	1/3	4/5	1/3	1/3	1/3	1/3	1/3	1/3	1/2	1/3
3	1/2	1/2	3	1/2	1/2	1/3	1/3	1/3	1/2	1/3	1/2	1/3	1	1/2	1
4	1/2	4/3	1	1/2	2	1/2	1/2	1	1/2	1/2	2	2	1/2	1/2	1/2
5	1/2	1	1/2	1/2	1/2	1/2	1	1/2	1/2	1/2	1/2	1/2	1/2	1/3	1
BE1	1	1/3	1/2	1/2	1	1	1	1	1/2	1/2	1/2	2/3	4	1/2	3
2	1	3	1/2	1/2	1	1/2	1/3	1/2	1/3	1/2	1/2	1/2	1/2	1/2	1/3
3	2/3	2/3	1	2	2/3	1/2	2	1/2	1/3	1	1	1/2	1	1/2	1/2
4	1	1	1	1	1	1	1	1	1	1/2	1/2	1	1	1	1
5	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1	1/2	1/2	1/2	1/2	1/2	1/2	1/2
SOD1	1	3/4	1	1/3	1	1/2	1	1/2	1/3	3	3	4	1	1	1
2	4	1/2	1	3	3	3/4	1	3	3	3	3	3/4	1	3/4	1
3	1/3	1/2	1/3	1/2	1/2	1/2	1/2	1/2	1/3	1/2	1/2	1/2	1/2	1/2	1/2
ACP1	1/2	1/2	1	1/3	1	1/2	1/2	1/3	1/2	1/2	1/2	1/2	1/2	1/4	1/2
2	1/2	1	1	2	1	1	1/2	1	1/2	2	2	1/2	1/2	4	1/2
3	1	1	0	1/3	1	1	1/2	1	1/3	1/2	1/3	1	1/3	1/4	1/4
GDH1	1	2	1	1	1/2	1/2	1	1	1	1/2	1/3	1/2	1	1/2	1/2
LTD1	1/4	1/3	1/2	1	1/2	1/3	1/2	1/3	1/4	1	1	1	1/2	3/4	1/2
2	1/2	1/3	1/4	1	1/3	1/3	2/3	1/4	1/2	1/2	2	1/2	0	1/2	0
LDH1	1/3	1/3	1/2	2	1	1/2	1	1/2	1/2	1/2	1	1/2	1/2	1/3	1/3
2	1/2	1	1/4	1	1/3	1/3	1/3	1/3	3	2	2	1/4	1/4	1/4	1/2
PGM1	1/4	1/3	1	1	1/2	1/3	1/3	2/3	2	1	1	1	1	1/2	2
2	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
3	1/2	2/3	2/3	2/3	1/2	1	1/2	1	2/3	3	3	1	2	1/2	2

TABLE E-3. Allozyme gel data, ground site, 1993. See Table E-1 for details.

LOCI	22	24	27	30	33	42	43	44	45	46	47	48	49	57	63
AE1	2	4/5	0	1/2	2	1/5	3/6	1	1/3	1/5	1	1/5	2	1	1
2	1/2	1/2	1	1/2	1/2	1	1/2	1/3	1/3	1/2	2	1	1	1/3	1/2
3	1/2	1/3	3	4/5	1/2	4/5	4/5	1	1	1	1	1/3	4	1	1/4
4	1/2	1/2	1	1/2	1/2	4/5	5	2/4	2/4	2/4	1/2	1/2	3/4	1/3	1/3
PE1	3	3	2	1/3	1/2	2	1/2	1	1/2	2	2	1/6	1	1	1
2	1/3	1/3	1/3	1/3	1	1	3	1/3	1/3	1	1	2	1/3	2	1
3	1/3	2/3	2	1/3	1/2	1/3	3	1	1/2	3	3	2/3	1/3	4	4
4	2	1/2	1	1/2	1/3	1/3	1	1/2	1/2	1/2	1/3	1/2	3/4	1	1/2
5	1	2	2	1/2	1/2	2	2	2	1	2	2	1/2	1/2	1/2	2
BE1	1/2	2	1	1/2	2/3	1/2	3	3/4	1/2	1	1	1	1	1/2	1/2
2	1/2	1/2	1/2	1/2	1	1/2	1/2	1/2	1/2	1/2	1	1	1/2	1	1
3	1	1/2	1/2	1	1/2	3	2	3	1/2	1	1/2	1/3	3	2	2
4	1	1	1/2	1/2	1/2	1/4	1	2	1	1	1/4	1/3	1/4	1/2	1/2
5	1/2	1	1	1	1/2	1	2	1/2	2	1/2	1	1/2	1	1/2	1/2
SOD1	1/2	1/2	1	3	1/4	3	3	1/2	1/2	1/2	3/4	3	3	1/2	1/3
2	3/4	3	4	3	3	4	4	4	1/2	4	4	1	1	3	1/3
3	1/3	1/3	1	1/2	1/3	1	1/3	1/3	1/2	1/3	1/3	1	1	1/2	1/2
ACP1	1/3	1/2	1/2	1/2	1/3	1/2	2/3	2/4	1/2	1/2	1/2	1/2	1/2	1/2	1/2
2	1/2	1/4	1/3	2	3	3	3	3	1	3	3	1	1	1/3	1/3
3	2	1/3	1/2	1/2	2	2	2	2	1	2	2	1/2	1/2	1	1/2
GDH1	1	1/4	1/4	1/2	1/3	1/3	1/4	1/3	1/2	1/3	1/5	2	1	1	1
LTD1	1/2	1/4	2	1	1	2	0	1	1/2	3	2	2	1	1/3	1/2
2	2	2	2	1	2/3	2	1	1	1/2	2	2	1/2	1/2	1/3	1/2
LDH1	2	1	1	1/2	2	1	2	2	1/3	3	2/3	1/2	1/2	1/2	1/2
2	1	1	1	1	1/2	1	1	1	1	1	1	1/4	2	2	
PGM1	1	1/2	1/4	1	1/2	2	1/2	1/2	1/4	1/2	2	2	1	1	1
2	1	1	1	1	1	1	1	1	1	1	1	1/2	1/2	1	1
3	2	1/2	1	3	1/3	1	1	1	1/2	1	1	1/2	1	2	2

TABLE E-3. cont'd (ground site).

LOCI	64	65	93	105	106	108	109	110	111	112	113	115	116	117	122
AE1	1	1/2	1/3	4/5	4/5	1/2	1/3	4/5	1/2	3/4	4	1/4	1/5	1/4	1/5
2	1/2	1/2	2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	2/3	1/2	2/3
3	1/3	1/3	1/3	1/2	1/2	2/3	1/2	1	1	1/2	1/2	1	3/4	1/3	4
4	3/4	1/2	1/2	2/4	2/4	1/2	1/2	1/4	1/2	2/4	3/4	1/2	1/2	1/2	1/2
PE1	3/4	2/3	1/6	7/8	7	2	3/4	1/2	2/3	1/3	1	5/6	9/6	5/6	9/6
2	1/2	1/3	1/2	1/3	1/3	1/3	1/3	1/3	1/3	1/3	1/3	1/3	1/3	1/3	1/3
3	3	1/2	1/3	1/3	1/2	1	1/3	1/2	1/3	1	1	1/3	1/2	1/3	1/2
4	1/3	1/3	1/2	2	2	1/2	1/2	1/2	2	2	2	1/2	1/2	1/2	1/2
5	1/2	1/2	1/2	1/2	1/2	1	1	1/2	1/2	2	2	1/2	1/2	1/2	1/2
BE1	1/2	1/2	2/3	1	3	4	2/3	4	1/2	2/3	2	1/2	1/2	1/2	1/2
2	1/2	1/2	1	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1	1/2	1
3	1/2	1/2	2	1	1	1	2	1/2	2	1/2	1/2	2	2/3	2	2/3
4	1	1/3	1	1	1	1	1	1	1	1	1	1	1	1	1
5	1/2	1/2	1/2	1/2	1/2	1/2	1	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2
SOD1	1/3	1/3	1/2	1/2	1/2	1	1/2	3/4	1/3	3/4	3	1/3	1	1/2	1
2	1	3	3/4	3	3	3/4	1/3	3	3/4	3	3	3/4	3/4	3/4	3/4
3	1/4	1/2	1/2	1/2	1/2	1/2	1/3	1/2	1/2	1/2	1/2	1/2	1/2	1/2	1/2
ACP1	1/2	1/3	1/2	1/3	1/3	1/2	1/2	1/2	1/3	1/2	1/2	1/3	1/2	1/3	1/2
2	1/2	3	1/2	1/3	1/2	1/2	1/2	1/2	3	2	2	3	1	3	2/3
3	1/3	1/3	3/4	1	1	1/3	1/4	1	1/3	1	1	1/3	1	1/3	1
GDH1	1/5	1/4	1	1	1	1	1/2	1/2	1	2	2	1	4	1	1/4
LTD1	1/3	1	1	1/4	1	1/2	1	1/2	1	1	1/4	1	1/4	1	1/4
2	1/3	2	1/2	2	2	0	1/2	1/2	1	1/2	1/2	3	1/2	1	1/2
LDH1	1/3	2	1/2	1/2	1/2	1	1/2	1/2	1	1/2	1	2	1	0	1/3
2	3	1/3	1/4	1/2	1/2	1/3	1	1/4	1	1/4	1/2	1	1/2	1/2	1/3
PGM1	1	1	2	1/2	1/2	2	1	1/3	1	1/3	1/2	1	2	1	2
2	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
3	1/2	2/4	2	1	1	2	1	1	1	1	1	1	2/3	1	1/2

TABLE E-4. Nei's genetic distance values for control site, 1993.
 Nei's D (genetic distance) is above the diagonal, I (genetic identity) is below the diagonal and J(X) (average homozygosity) is on the diagonal.

	6	7	9	10	14	19
6	(.723)	.647	.514	.585	.351	.499
7	.524	(.643)	.595	.573	.647	.529
9	.598	.552	(.652)	.446	.536	.237
10	.557	.564	.64	(.688)	.68	.281
14	.704	.524	.585	.506	(.723)	.66
19	.607	.589	.789	.755	.517	(.661)
21	.625	.635	.577	.627	.625	.573
23	.681	.663	.6	.67	.681	.639
25	.521	.591	.651	.622	.521	.596
28	.485	.501	.779	.671	.436	.723
32	.626	.429	.556	.503	.736	.488
38	.528	.56	.596	.748	.554	.711
39	.589	.639	.634	.591	.589	.671
40	.659	.575	.558	.649	.685	.568
41	.535	.446	.806	.627	.523	.747
55	.347	.559	.53	.479	.335	.476
56	.407	.534	.517	.393	.407	.401
58	.637	.46	.618	.458	.625	.613
66	.555	.616	.653	.583	.581	.581
72	.539	.572	.568	.479	.623	.564
77	.448	.376	.422	.373	.559	.41
88	.568	.616	.585	.649	.581	.635
89	.503	.427	.57	.735	.44	.684
81	.57	.591	.69	.684	.594	.609
82	.593	.545	.556	.527	.686	.635
83	.637	.563	.588	.531	.598	.57
84	.62	.658	.639	.623	.53	.581
85	.563	.528	.579	.524	.576	.603
86	.629	.611	.538	.604	.681	.575
87	.642	.625	.703	.645	.655	.644

TABLE E-4 cont'd (control site).

	21	23	25	28	32	38
6	.471	.385	.651	.724	.469	.638
7	.454	.411	.525	.69	.845	.579
9	.549	.511	.429	.25	.587	.517
10	.466	.401	.475	.398	.686	.29
14	.471	.385	.651	.829	.306	.591
19	.556	.447	.517	.324	.718	.341
21	(.679)	.202	.469	.574	.519	.56
23	.817	(.571)	.488	.557	.499	.569
25	.626	.614	(.75)	.48	.658	.633
28	.563	.573	.619	(.75)	.73	.462
32	.595	.607	.518	.482	(.732)	.644
38	.571	.566	.531	.63	.525	(.696)
39	.852	.795	.591	.553	.612	.56
40	.827	.857	.596	.545	.578	.54
41	.566	.574	.563	.788	.583	.61
55	.495	.499	.518	.506	.464	.415
56	.495	.499	.482	.482	.464	.415
58	.5	.531	.576	.551	.798	.571
66	.827	.697	.66	.621	.578	.553
72	.73	.701	.494	.553	.548	.525
77	.491	.619	.549	.375	.417	.38
88	.667	.639	.609	.495	.591	.605
89	.533	.51	.593	.581	.488	.628
81	.788	.709	.643	.619	.602	.581
82	.735	.697	.583	.557	.577	.618
83	.59	.613	.509	.469	.581	.568
84	.773	.756	.609	.558	.591	.54
85	.568	.575	.514	.489	.495	.614
86	.73	.722	.566	.45	.625	.574
87	.811	.751	.604	.579	.586	.6

TABLE E-4 cont'd (control site).

	39	40	41	55	56	58
6	.529	.417	.625	1.057	.898	.451
7	.448	.553	.807	.581	.628	.777
9	.455	.584	.216	.635	.659	.482
10	.526	.432	.466	.736	.933	.782
14	.529	.379	.649	1.092	.898	.471
19	.399	.566	.292	.742	.913	.489
21	.161	.19	.57	.704	.704	.693
23	.229	.154	.556	.696	.696	.634
25	.525	.517	.574	.658	.729	.552
28	.593	.606	.238	.681	.729	.597
32	.492	.549	.54	.767	.767	.226
38	.579	.617	.494	.879	.879	.56
39	(.643)	.213	.52	.604	.677	.566
40	.808	(.661)	.556	.796	.796	.734
41	.595	.573	(.679)	.809	.809	.547
55	.546	.451	.445	(.768)	.111	.704
56	.508	.451	.445	.895	(.768)	.679
58	.568	.48	.579	.495	.507	(.679)
66	.753	.77	.627	.526	.476	.533
72	.699	.74	.618	.477	.477	.532
77	.524	.605	.395	.29	.326	.404
88	.671	.595	.48	.526	.426	.533
89	.6	.54	.571	.427	.33	.52
81	.823	.799	.651	.541	.459	.526
82	.685	.69	.599	.422	.461	.613
83	.634	.584	.562	.49	.554	.548
84	.74	.676	.547	.501	.514	.493
85	.625	.548	.581	.407	.496	.541
86	.694	.699	.5	.407	.419	.514
87	.833	.808	.635	.47	.445	.527

TABLE E-4 cont'd (control site).

	66	72	77	88	89	81
6	.588	.618	.804	.565	.687	.562
7	.484	.559	.979	.484	.851	.525
9	.426	.566	.862	.536	.562	.372
10	.54	.736	.986	.432	.307	.38
14	.543	.473	.581	.543	.82	.521
19	.543	.573	.892	.454	.379	.496
21	.19	.315	.711	.405	.63	.238
23	.36	.355	.48	.447	.674	.344
25	.416	.705	.599	.496	.523	.442
28	.476	.592	.98	.704	.544	.48
32	.549	.602	.875	.527	.718	.507
38	.593	.644	.967	.502	.465	.544
39	.283	.358	.646	.399	.51	.195
40	.261	.302	.503	.52	.617	.224
41	.467	.481	.93	.734	.56	.43
55	.642	.741	1.239	.642	.85	.614
56	.742	.741	1.122	.853	1.11	.779
58	.629	.631	.906	.629	.655	.643
66	(.661)	.372	.823	.497	.617	.148
72	.689	(.768)	.876	.666	.909	.417
77	.439	.416	(1.268)	.605	.895	.781
88	.608	.514	.546	(.661)	.502	.397
89	.54	.403	.409	.605	(.696)	.462
81	.862	.659	.458	.672	.63	(.75)
82	.662	.729	.518	.662	.524	.544
83	.57	.477	.471	.57	.609	.6
84	.676	.602	.449	.649	.487	.71
85	.548	.521	.475	.575	.587	.604
86	.603	.572	.623	.849	.587	.63
87	.849	.699	.465	.658	.534	.887

TABLE E-4 cont'd (control site).

	82	83	84	85	86	87
6	.522	.45	.478	.574	.464	.444
7	.606	.574	.419	.639	.492	.47
9	.588	.532	.447	.546	.62	.352
10	.64	.633	.474	.647	.504	.439
14	.377	.515	.636	.551	.384	.424
19	.455	.563	.543	.506	.553	.44
21	.308	.528	.257	.566	.315	.209
23	.361	.49	.28	.554	.326	.286
25	.54	.676	.496	.665	.57	.504
28	.586	.756	.583	.716	.798	.547
32	.551	.543	.527	.704	.47	.535
38	.481	.565	.617	.488	.555	.51
39	.378	.456	.301	.47	.365	.182
40	.371	.539	.392	.601	.359	.213
41	.513	.576	.604	.543	.693	.454
55	.862	.714	.69	.9	.9	.755
56	.775	.59	.666	.702	.869	.81
58	.49	.601	.706	.615	.666	.64
66	.412	.563	.392	.601	.506	.163
72	.315	.741	.508	.652	.559	.358
77	.658	.752	.801	.745	.473	.766
88	.412	.563	.433	.553	.163	.419
89	.646	.496	.719	.532	.532	.628
81	.609	.511	.342	.504	.462	.12
82	(.634)	.542	.522	.463	.281	.441
83	.582	(.625)	.426	.119	.456	.574
84	.593	.653	(.661)	.506	.378	.339
85	.629	.887	.603	(.643)	.448	.563
86	.755	.634	.685	.639	(.643)	.427
87	.643	.563	.712	.569	.653	(.643)

TABLE E-5. Nei's genetic distance values for antenna site, 1993. Nei's D (genetic distance) is above the diagonal, I (genetic identity) is below the diagonal and J(X) (average homozygosity) is on the diagonal.

94	2	3	4	11	12
94 (.723)	.368	.365	.675	.431	.352
2 .692	(.696)	.158	.438	.303	.351
3 .694	.854	(.643)	.378	.316	.365
4 .509	.646	.685	(.714)	.638	.669
11 .65	.739	.729	.528	(.705)	- .195
12 .703	.704	.694	.512	.822	(.75)
13 .547	.722	.619	.55	.642	.647
26 .484	.675	.689	.551	.632	.551
31 .788	.684	.726	.559	.719	.812
36 .51	.597	.608	.526	.555	.576
50 .491	.632	.603	.598	.628	.571
51 .54	.613	.573	.519	.634	.651
52 .454	.538	.521	.482	.485	.422
54 .519	.591	.589	.584	.588	.558
59 .735	.761	.754	.631	.769	.722
60 .546	.716	.676	.602	.619	.6
61 .502	.484	.504	.451	.619	.666
70 .528	.615	.587	.544	.586	.63
71 .654	.628	.654	.633	.713	.704
75 .559	.671	.672	.687	.755	.708
97 .557	.581	.662	.574	.715	.666
98 .497	.584	.581	.526	.671	.601
99 .554	.59	.667	.557	.701	.642
100 .691	.65	.662	.561	.686	.639
101 .827	.724	.767	.585	.732	.71
104 .778	.73	.746	.547	.763	.715
118 .542	.605	.63	.572	.719	.66
119 .535	.597	.608	.551	.607	.613
120 .514	.651	.678	.573	.676	.614
121 .491	.566	.616	.507	.628	.571

TABLE E-5 cont'd (control site).

	13	26	31	36	50	51
94	.604	.725	.238	.674	.712	.616
2	.326	.392	.379	.515	.459	.49
3	.479	.372	.32	.497	.506	.558
4	.598	.595	.582	.643	.514	.657
11	.444	.458	.329	.589	.466	.456
12	.436	.597	.209	.552	.561	.43
13	(.714)	.528	.451	.465	.493	.482
26	.59	(.679)	.68	.72	.511	.608
31	.637	.507	(.661)	.556	.543	.484
36	.628	.487	.573	(.679)	.654	.608
50	.611	.6	.581	.52	(.661)	.181
51	.617	.545	.616	.545	.834	(.732)
52	.605	.583	.462	.57	.603	.573
54	.683	.523	.607	.574	.517	.466
59	.534	.572	.681	.448	.58	.611
60	.589	.51	.558	.604	.544	.543
61	.587	.476	.625	.476	.525	.58
70	.671	.481	.658	.507	.54	.6
71	.595	.481	.684	.636	.566	.538
75	.725	.603	.663	.603	.637	.691
97	.601	.519	.667	.617	.611	.621
98	.705	.539	.52	.487	.573	.57
99	.544	.52	.605	.571	.566	.563
100	.535	.521	.597	.494	.528	.528
101	.598	.507	.851	.56	.581	.578
104	.621	.561	.788	.548	.542	.54
118	.572	.507	.689	.6	.568	.578
119	.68	.487	.587	.553	.533	.532
120	.629	.531	.581	.731	.465	.483
121	.624	.52	.527	.467	.5	.462

TABLE E-5 cont'd (control site).

	52	54	59	60	61	70
94	.79	.657	.308	.605	.689	.638
2	.621	.525	.273	.335	.725	.486
3	.653	.529	.282	.392	.685	.532
4	.731	.538	.461	.508	.797	.608
11	.725	.532	.263	.48	.48	.534
12	.863	.584	.326	.51	.406	.462
13	.503	.381	.628	.53	.532	.399
26	.54	.649	.558	.673	.741	.733
31	.772	.499	.384	.584	.47	.418
36	.562	.556	.803	.504	.741	.68
50	.505	.66	.545	.608	.643	.617
51	.557	.763	.493	.611	.545	.511
52	(.732)	.712	.928	.852	1.01	.621
54	.491	(.723)	.705	.558	.932	.504
59	.395	.494	(.759)	.605	.713	.593
60	.427	.572	.546	(.652)	.61	.473
61	.364	.394	.49	.543	(.598)	.67
70	.538	.604	.553	.623	.512	(.696)
71	.413	.654	.614	.636	.526	.628
75	.506	.696	.679	.68	.587	.62
97	.472	.584	.636	.643	.582	.692
98	.405	.548	.622	.658	.603	.623
99	.45	.541	.639	.583	.484	.654
100	.436	.544	.648	.643	.526	.568
101	.475	.568	.706	.544	.483	.592
104	.466	.568	.747	.559	.462	.516
118	.488	.633	.656	.571	.54	.579
119	.418	.65	.56	.618	.49	.714
120	.4	.708	.57	.688	.551	.58
121	.372	.581	.517	.639	.611	.579

TABLE E-5 cont'd (control site).

71	75	97	98	99	100
94 .424	.582	.586	.699	.591	.37
2 .465	.399	.543	.537	.528	.431
3 .425	.398	.412	.543	.405	.412
4 .457	.375	.556	.643	.585	.578
11 .338	.281	.336	.399	.356	.377
12 .351	.346	.406	.51	.443	.448
13 .519	.322	.509	.349	.608	.626
26 .733	.506	.657	.617	.655	.652
31 .379	.411	.404	.654	.502	.515
36 .452	.506	.484	.72	.56	.706
50 .569	.451	.493	.556	.569	.639
51 .621	.369	.477	.562	.575	.639
52 .885	.681	.75	.903	.798	.831
54 .424	.363	.538	.601	.614	.608
59 .488	.387	.452	.475	.448	.434
60 .452	.385	.441	.419	.539	.441
61 .643	.532	.541	.507	.725	.643
70 .465	.478	.369	.473	.425	.565
71 (.696)	.399	.389	.412	.405	.431
75 .671	(.714)	.25	.331	.344	.383
97 .678	.779	(.598)	.376	.154	.489
98 .662	.718	.687	(.679)	.473	.528
99 .667	.709	.858	.623	(.696)	.474
100 .65	.682	.613	.59	.623	(.625)
101 .724	.598	.611	.52	.553	.667
104 .692	.621	.557	.561	.491	.637
118 .724	.663	.767	.613	.671	.542
119 .688	.641	.617	.75	.558	.59
120 .651	.699	.672	.632	.637	.598
121 .566	.663	.568	.707	.566	.597

TABLE E-5 cont'd (control site).

	101	104	118	119	120	121
94	.19	.251	.612	.625	.665	.712
2	.323	.315	.502	.515	.429	.569
3	.265	.293	.462	.497	.389	.484
4	.536	.604	.559	.595	.557	.679
11	.311	.271	.329	.5	.392	.466
12	.342	.335	.416	.489	.487	.561
13	.514	-.476	.559	.386	.464	.472
26	.68	.578	.68	.72	.633	.654
31	.161	.238	.372	.533	.542	.641
36	.58	.601	.511	.593	.313	.762
50	.543	.612	.566	.629	.766	.693
51	.549	.616	.549	.631	.727	.772
52	.744	.763	.718	.872	.916	.988
54	.565	.566	.457	.431	.345	.543
59	.348	.291	.422	.58	.562	.66
60	.608	.581	.56	.482	.374	.447
61	.728	.773	.617	.712	.597	.493
70	.524	.662	.546	.336	.544	.546
71	.323	.368	.323	.373	.429	.569
75	.514	.476	.411	.444	.358	.411
97	.493	.586	.265	.484	.397	.565
98	.654	.578	.489	.288	.459	.347
99	.593	.712	.399	.583	.451	.569
100	.405	.45	.613	.528	.514	.515
101	(.661)	.115	.412	.511	.542	.641
104	.891	(.723)	.417	.512	.539	.66
118	.662	.659	(.661)	.467	.447	.615
119	.6	.599	.627	(.679)	.483	.292
120	.581	.584	.64	.617	(.572)	.402
121	.527	.517	.541	.747	.669	(.661)

TABLE E-6. Nei's genetic distance values for ground site, 1993.
 Nei's D (genetic distance) is above the diagonal, I (genetic identity) is below the diagonal and J(X) (average homozygosity) is on the diagonal.

22	24	27	30	33	42
22 (.75)	.41	.681	.405	.397	.813
24 .663	(.679)	.408	.473	.556	.598
27 .506	.665	(.804)	.715	.641	.352
30 .667	.623	.489	(.696)	.48	.702
33 .672	.573	.527	.619	(.661)	.629
42 .444	.55	.703	.496	.533	(.821)
43 .449	.447	.511	.501	.539	.593
44 .523	.431	.473	.555	.667	.576
45 .63	.608	.584	.56	.507	.455
46 .598	.592	.644	.477	.588	.703
47 .529	.544	.652	.479	.623	.763
48 .5	.526	.589	.532	.429	.594
49 .529	.47	.511	.623	.501	.607
57 .471	.532	.455	.537	.564	.326
63 .549	.577	.542	.608	.624	.501
64 .522	.59	.479	.582	.445	.449
65 .704	.686	.592	.663	.75	.486
93 .626	.685	.469	.583	.612	.451
105 .701	.711	.556	.597	.613	.514
106 .671	.693	.507	.595	.637	.443
108 .663	.607	.569	.573	.549	.493
109 .702	.725	.555	.702	.626	.464
110 .574	.672	.592	.663	.597	.511
111 .667	.701	.573	.705	.592	.519
112 .568	.662	.442	.718	.579	.449
113 .482	.668	.443	.684	.514	.506
115 .597	.641	.553	.606	.649	.511
116 .501	.716	.609	.547	.521	.491
117 .587	.644	.568	.623	.626	.476
122 .525	.677	.584	.586	.56	.565

TABLE E-6 cont'd (control site).

	43	44	45	46	47	48
22	.802	.648	.462	.514	.637	.693
24	.804	.843	.497	.523	.608	.643
27	.671	.75	.538	.439	.427	.529
30	.691	.589	.579	.739	.736	.632
33	.618	.406	.679	.531	.472	.847
42	.522	.551	.788	.352	.27	.52
43	(.804)	.503	.805	.549	.533	.803
44	.604	(.821)	.662	.522	.581	.814
45	.447	.516	(.643)	.538	.746	.458
46	.578	.593	.584	(.804)	.204	.68
47	.587	.559	.474	.815	(.839)	.551
48	.448	.443	.632	.507	.577	(.714)
49	.466	.562	.546	.443	.445	.603
57	.398	.495	.623	.409	.4	.494
63	.483	.536	.606	.495	.554	.537
64	.441	.399	.634	.567	.505	.548
65	.491	.523	.606	.542	.555	.468
93	.518	.525	.634	.506	.567	.55
105	.435	.478	.676	.665	.544	.526
106	.424	.49	.659	.554	.438	.462
108	.356	.469	.676	.51	.487	.503
109	.543	.549	.759	.531	.507	.537
110	.504	.523	.732	.542	.53	.575
111	.561	.59	.707	.585	.549	.481
112	.489	.531	.641	.477	.432	.481
113	.455	.472	.61	.455	.412	.506
115	.577	.57	.645	.613	.588	.446
116	.41	.442	.667	.46	.474	.593
117	.592	.598	.648	.592	.543	.432
122	.495	.515	.653	.558	.559	.538

TABLE E-6 cont'd (control site).

49	57	63	64	65	93
22 .636	.754	.6	.651	.351	.469
24 .755	.631	.55	.528	.378	.379
27 .67	.788	.612	.737	.524	.758
30 .474	.621	.498	.541	.411	.539
33 .69	.573	.472	.811	.287	.491
42 .499	.1.121	.691	.802	.722	.795
43 .764	.922	.727	.819	.711	.657
44 .576	.704	.623	.919	.647	.645
45 .604	.474	.501	.456	.501	.455
46 .814	.894	.703	.567	.613	.682
47 .81	.915	.591	.682	.589	.567
48 .506	.705	.621	.602	.76	.599
49 (.768)	.791	.547	.545	.663	.817
57 .453	(.768)	.17	.48	.501	.421
63 .579	.844	(.714)	.465	.444	.385
64 .58	.619	.628	(.625)	.511	.508
65 .516	.606	.641	.6	(.625)	.463
93 .442	.656	.68	.602	.63	(.652)
105 .557	.532	.564	.535	.672	.591
106 .506	.494	.525	.494	.641	.628
108 .412	.51	.516	.565	.619	.711
109 .593	.593	.602	.685	.671	.726
110 .567	.58	.601	.557	.643	.616
111 .586	.586	.633	.65	.69	.689
112 .525	.476	.544	.528	.609	.623
113 .523	.512	.591	.528	.528	.568
115 .516	.614	.612	.627	.722	.654
116 .432	.585	.606	.521	.592	.731
117 .517	.644	.654	.63	.713	.699
122 .48	.571	.619	.561	.633	.733

TABLE E-6 cont'd (control site).

105	106	108	109	110	111
22 .355	.399	.411	.353	.555	.405
24 .342	.367	.5	.322	.398	.355
27 .587	.68	.563	.588	.524	.557
30 .515	.519	.556	.353	.411	.349
33 .489	.451	.599	.469	.515	.524
42 .665	.814	.708	.769	.672	.655
43 .832	.857	1.033	.611	.685	.578
44 .737	.714	.757	.599	.647	.527
45 .392	.417	.391	.276	.311	.346
46 .408	.591	.673	.634	.613	.536
47 .608	.825	.719	.679	.635	.6
48 .643	.771	.687	.623	.554	.732
49 .586	.681	.886	.522	.567	.534
57 .631	.705	.674	.522	.545	.534
63 .572	.644	.662	.508	.509	.457
64 .626	.704	.571	.378	.585	.431
65 .398	.444	.48	.398	.442	.371
93 .526	.465	.341	.32	.485	.372
105 (.679)	.108	.458	.379	.319	.355
106 .898	(.714)	.444	.329	.29	.399
108 .632	.642	(.705)	.48	.397	.431
109 .685	.72	.619	(.652)	.318	.213
110 .727	.748	.672	.727	(.625)	.332
111 .701	.671	.65	.808	.717	(.696)
112 .701	.76	.535	.755	.826	.705
113 .693	.699	.534	.656	.773	.672
115 .693	.637	.564	.747	.667	.839
116 .635	.606	.676	.621	.592	.614
117 .712	.654	.566	.767	.657	.848
122 .663	.633	.677	.648	.619	.654

TABLE E-6 cont'd (control site).

112	113	115	116	117	122
22 .565	.729	.516	.69	.532	.644
24 .412	.404	.445	.333	.439	.391
27 .817	.814	.593	.496	.566	.538
30 .331	.38	.5	.603	.473	.534
33 .546	.666	.432	.653	.469	.58
42 .802	.681	.672	.71	.743	.571
43 .715	.788	.551	.892	.524	.703
44 .633	.75	.562	.816	.514	.664
45 .445	.494	.439	.405	.433	.427
46 .739	.788	.49	.777	.524	.583
47 .839	.888	.532	.746	.61	.582
48 .732	.681	.808	.523	.84	.619
49 .644	.648	.661	.839	.659	.733
57 .742	.67	.487	.537	.441	.56
63 .608	.526	.492	.501	.424	.479
64 .639	.638	.468	.652	.463	.578
65 .496	.638	.326	.525	.338	.457
93 .473	.566	.425	.313	.359	.311
105 .355	.367	.367	.454	.34	.411
106 .275	.358	.451	.501	.424	.458
108 .625	.628	.572	.391	.569	.39
109 .281	.421	.292	.477	.265	.434
110 .192	.257	.404	.525	.419	.48
111 .349	.398	.176	.488	.165	.424
112(.696)	.115	.458	.603	.452	.511
113 .891	(.768)	.507	.537	.501	.494
115 .632	.602	(.688)	.597	.098	.439
116 .547	.585	.551	(.643)	.499	.145
117 .636	.606	.907	.607	(.652)	.391
122 .6	.61	.645	.865	.676	(.616)